

JOURNAL
OF THE
Elisha Mitchell Scientific Society

VOLUME 64
1948



This Society was founded in 1883 through the efforts of
Francis Preston Venable

Published for the Society
by the
University of North Carolina Press

CONTENTS OF VOLUME 64

LIMNOLOGICAL OBSERVATIONS ON SOME STREAMS OF THE NEW RIVER WATERSHED IN THE VICINITY OF MOUNTAIN LAKE, VIRGINIA. <i>Charles S. Shoup</i>	1
A COMPARATIVE STUDY OF ZYGOTE GERMINATION IN THE SAPROLEGNACEAE. <i>Arthur William Ziegler</i>	13
THE BASKING SHARK, <i>Cetorhinus maximus</i> , ON THE NORTH CAROLINA COAST. <i>E. W. Gudger</i>	41
THE VASCULAR FLORA OF THE OLIVINE DEPOSITS OF NORTH CAROLINA AND GEORGIA. <i>Albert E. Radford</i>	45
THE GENUS <i>CODIUM</i> IN NORTH CAROLINA. <i>Louis G. Williams</i>	107
A CYTOLOGICAL STUDY OF THE EFFECTS OF COLCHICINE, INDOLE-3-ACETIC ACID, POTASSIUM CYANIDE, AND 2,4-D ON PLANT CELLS. <i>Alice G. Ryland</i>	117
A SECOND REPORT ON PARASITIC COPEPODS COLLECTED AT BEAUFORT, N. C. <i>A. S. Pearse</i>	127
A NEW MEMBER OF THE PLASMODIOPHORACEAE. <i>William R. Pendergrass</i> ..	132
NOTES ON SOME HIGHER FUNGI. <i>W. C. Coker</i>	135
PROCEEDINGS OF THE FORTY-FIFTH ANNUAL MEETING OF THE NORTH CAROLINA ACADEMY OF SCIENCE.....	147
PROCEEDINGS OF THE ELISHA MITCHELL SCIENTIFIC SOCIETY.....	182
OBSERVATIONS ON THE HELMINTHS AND ECTOPARASITES OF THE COTTON RAT, <i>Sigmodon hispidus hispidus</i> SAY AND ORD, IN GEORGIA AND NORTH CAROLINA. <i>Reinard Harkema</i>	183
A COMPARATIVE STUDY OF THE SUSCEPTIBILITY OF GUINEA PIGS AND HAMSTERS TO AN INFECTION WITH PIG ASCARIS. <i>John E. Larsh, Jr., and Margaret S. Gravatt</i>	192
FURTHER STUDIES IN MICE ON THE EFFECT OF ALCOHOL ON ACQUIRED IMMUNITY TO <i>Trichinella spiralis</i> . <i>John E. Larsh, Jr., and Orlin K. Fletcher, Jr.</i>	196
A SURVEY FOR INTESTINAL PROTOZOA AMONG FOOD HANDLERS OF A UNIVERSITY DINING HALL. <i>John E. Larsh, Jr., Margaret S. Gravatt, and Hazel B. Gilchrist</i>	204
COMMON ECTOPARASITES OF THE NORWAY RAT, <i>Rattus norvegicus</i> (ERXL.), IN NORTH CAROLINA AND THE EFFECTS OF 10 PER CENT DDT DUST TREATMENTS IN THEIR CONTROL AND UPON THE SEROLOGY OF THE RAT. <i>H. F. Schoof</i>	208
REACTIONS OF THE ADRENAL CORTEX TO DIPHTHERIA TOXIN. <i>John Nichols</i>	216
THE TIGER SHARK, <i>Galeocерdo tigrinus</i> , ON THE NORTH CAROLINA COAST AND ITS FOOD AND FEEDING HABITS THERE. <i>E. W. Gudger</i>	221
A LIST OF CALANOID COPEPODS FROM THE PLANKTON AT BEAUFORT, N. C. <i>W. H. Sutcliffe, Jr.</i>	234
NOTES ON THE DISTRIBUTION OF <i>Carya aquatica</i> IN NORTH CAROLINA. <i>William B. Fox</i>	237

STUDIES IN THE COMPOSITAE OF NORTH CAROLINA. I. <i>Liatris</i> . R. K. Godfrey	241
THE COASTAL PLAIN OF NORTH CAROLINA. H. W. Straley, III	250
SOUTHERN CHEMISTS: THEIR TRAINING AND CAREERS. Will S. DeLoach ..	253
A NEW SPECIES OF <i>ACHLYA</i> AND OF <i>DICTYUCHUS</i> . E. S. Beneke	261
STUDIES ON SOME WILD YEASTS FROM CHAPEL HILL AND VICINITY. Helen S. Sherwin	267
KERATINOPHILIC PHYCOMYCETES. I. A NEW GENUS OF THE SAPROLEGNIACEAE. Maeburn Bruce Huneycutt	277
NOTES ON CAROLINA FUNGI. W. C. Coker	287

JOURNAL
OF THE
Elisha Mitchell Scientific Society

Volume 64

June 1948

No. 1

LIMNOLOGICAL OBSERVATIONS ON SOME STREAMS OF THE
NEW RIVER WATERSHED IN THE VICINITY OF
MOUNTAIN LAKE, VIRGINIA

BY CHARLES S. SHOUP

*A contribution from the Mountain Lake Biological Station of the University of
Virginia, and Department of Biology, Vanderbilt University,
Nashville, Tennessee*

INTRODUCTION

In the summer of 1947, as a part of the work in limnology* at the Mountain Lake Biological Station, a survey of thermal and chemical characteristics of streams was undertaken preliminary to extensive investigations of the lake. Streams examined were those in the vicinity of Mountain Lake, on and adjacent to Salt Pond Mountain, Giles County. Some of the streams which were investigated are the same ones within Giles County previously examined with regard to fish distribution by Burton and Odum (1945), and may be identified on the U.S.G.S. (Virginia-West Virginia) Pearisburg Quadrangle map (Scale 1/62500), and on the U.S.G.S. (Virginia-West Virginia) Dublin Sheet (Scale 1/125000).

Salt Pond Mountain, on which Mountain Lake and the post office and hotel of the same name are located, rises to a height greater than 4000 ft. in northeastern Giles County. (Highest transit station is at Bald Knob, 4363 ft.) This represents a point on the divide between Atlantic and Mississippi drainage systems in Virginia, and the streams examined are all tributaries of New River, a stream originating in northwestern North Carolina which cuts across the parallel Appalachian ridges of Virginia to become the Kanawha of the Ohio drainage.

For investigation by the limnology group and because of accessibility, six streams of the above-mentioned drainage area were chosen. All stream-sampling localities lie within Giles County, with the exception of Station 3 at the headwaters of Sinking Creek, Craig County. (1) Sinking Creek, the most accessible stream examined, originates in a limestone spring of Craig County, and flows

* Personnel of the survey group: A. W. Burke (Va.); F. R. Derrick (N. C.); A. T. Hyde (Va.); C. T. Mann (Fla.); M. A. Miller (N. J.); J. Y. Peary (N. C.); J. B. Porter (Va.). C. S. Shoup, Party Leader. The author is indebted to these men for gathering data, their cooperation, and their close attention to details of operations.

westward between Sinking Creek Mountain and Gap Mountain and sinks underground in the cavernous limestone a few miles before entering New River. (2) Spruce Run Creek, a small lowland stream, originates also from a limey spring near Newport, Virginia, and flows in a narrow valley between Spruce Run Mountain and Gap Mountain, entering the New River near Goodwin's Ferry, Virginia. (3) Little Stony Creek, in contrast to the above, is a soft-water mountain stream originating on Salt Pond Mountain, from which it torrentially drops (3800 ft. to 1600 ft.) to the New River at Pembroke, Virginia. (4) Big Stony Creek, a typical mountain stream in its upper reaches, enters the lowland limestone near Kerns, and, gathering soluble buffers from limestone bedrock, quarries and cultivated lands, finally reaches the New River at Norcross not far below the bridge at Ripplemead. (5) Pond Drain, on top of Salt Pond Mountain, has all the characteristics of a small mountain brook, but in this instance it actually is, during most seasons, draining Mountain Lake, the high-altitude oligotrophic soft-water lake which lends its name to this section. (6) Hunter's Branch, also on top of Salt Pond Mountain, is a very small spring-fed brook whose chemical and thermal characteristics are typical of the highland streams of the region. Pond Drain and Hunter's Branch are tributaries of Little Stony Creek.

From an ecological viewpoint, these streams of mountain and lowland offer a favorable restricted locality for study because of their contrast in chemical and thermal characteristics and differences in biological productivity. In addition, the mountain and lowland streams reflect in the water chemistry the variation in solubilities of some constituents of their beds. In contrast, the higher streams flow over relatively insoluble sandstones and shales, while the lowland streams flow through more soluble dolomites and limestones with abundant materials sufficiently soluble to contribute a marked buffer action to the waters.

The stratigraphic situations which determine the relative amounts of soluble carbonate available as buffer in the bicarbonate buffer system operative in these streams, may be seen from the following geological descriptions (Butts, 1933) of the characteristic beds for each stream:

1. *Sinking Creek, Craig and Giles counties:*

- a. Upper: Cooper Ridge Dolomite (Ozarkian), a thick-bedded gray cherty dolomite of rather uniform character, with some interspersed sandstone.
- b. Lower: From above Maybrook downstream to New River: Stones River Dolomite of relatively pure limestone nature of the lower Ordovician (Canadian), a thick-bedded fossiliferous formation. These formations contribute varying amounts of calcium and magnesium carbonate to the stream.

2. *Spruce Run, Giles County:*

This small stream, throughout its length, meanders in a narrow valley with opportunity of passage over bedrock from both the Copper Ridge Dolomite and the Beekmantown formations. Abundant soluble carbonates contribute to the high alkalinity of this lowland (1662-2400 ft.) stream.

3. *Little Stony Creek, Giles County:*

In contrast with the above-mentioned streams, this one has its origin from

spring-fed mountain brooks at a high altitude on Salt Pond Mountain in the Clinch Sandstones (Silurian) of sediments and quartzites which make up the high ridges of the region. This is a hard white, grey, or reddish sandstone which contributes almost no buffer substances to the streams. In its lower portions, near Pembroke, this stream begins to pick up a higher bicarbonate alkalinity as it flows over thin alternating beds of broken dolomite of the Stones River and Beekmantown near New River.

4. *Big Stony Creek, Giles County:*

This stream, also originating in the Clinch Sandstone, is a typical mountain stream with characteristics of dissolved substances not unlike Little Stony Creek in its upper half. However, the lower half of this stream, from Foster Knob to New River at Norcross, traverses beds containing increasing quantities of the Beekmantown Dolomite with abundant commercial limestones. Drains and washes from adjacent quarries along the stream near its mouth add soluble carbonates.

5-6. *Pond Drain and Hunter's Branch, Giles County:*

These small streams are evidently confined to the Clinch Sandstone, and in their soft-water characters reflect the poor buffering action of this formation. Typically upland streams of variable flow, they filter through the broken sandstones and shales of this highland formation (3174-4000 ft.), with but small gain in buffer substances.

METHODS

The methods of stream examination and water analyses followed those previously reported upon for Tennessee (Shoup, 1940, 1944; Shoup, Peyton and Gentry, 1941), and were based upon those of Theroux, Eldridge and Mallmann (1936) for water analyses, and the New Hampshire biological survey report (Hoover et al., 1937) for most stream examination and grading. The main features of the methods are available in these publications, and special methods or references to them are cited below.

RESULTS

Stream Examination and Evaluation: The work of the survey group included not only field water analyses, but also some effort was made toward obtaining a general evaluation of the stream as a biologically-productive habitat, especially with reference to the presence of possible food and shelter for fishes. General field identification of bottom organisms and their comparative abundance, together with availability, yields some information on biological productivity. Bottom fauna were collected by the method suggested by Hazzard (1935) and extensively reviewed in the New Hampshire Report No. 2 (Hoover, et al., 1937). The procedure is accomplished by use of the Foerst square-foot sampler set up in at least three positions on characteristic stream bottom at a locality. Generally a sample is secured in the middle of a stream and in positions midway toward either bank. The apparatus is arranged with its bolting-cloth cone net down-stream, and then all stones are turned over and washed off from the square-

foot area marked off in front of the net, and all bottom sand and silt is stirred within the same square-foot area enclosed by the device. When the area has been thoroughly worked over, the contents swept into the net are dumped into a pan and thoroughly searched for organisms. Numbers of organisms and their total volume together constitute a food grade estimation. This is best shown by placing the specimens, freed of excess moisture, in a graduated centrifuge tube, and noting the total volume of material by displacement of water. In this instance, recognition must be made of numbers of organisms contributing to the total volume on the tripartite grading scale, viz:

Grade I (Exceptional richness). Volume greater than 2 cc. or 2 grams, with numbers greater than 50.

Grade II (Average). Volume from 1 to 2 cc. (1-2 grams), with more than 50 organisms.

Grade III (Poor). Volume less than 1 cc. (1 gram) and (or) not more than 50 organisms.

From the tabulations (Tables I-IV) it may be seen that the field observations and estimations favor the lowland streams in terms of both "wet food" volume and food grade. This is predominantly determined by numbers of organisms available rather than by distribution of species. It in no way detracts from the general consideration that warmer waters and more abundant microscopic forms with more rapid growing rates in the lowland streams contribute to the production of greater amounts of available food.

Temperature: Air and water temperatures were taken and recorded as °F. in the manner prescribed in the New Hampshire (1937) Report, and have previously been checked and recorded during the summer season by Burton and Odum (1945). Our temperature determinations ran somewhat higher than those indicated by Burton and Odum for Sinking Creek, but rather rapid flow during the season in Little Stony Creek and in Spruce Run accounted for slightly lower values for these localities. It would appear from our readings that water temperatures in mid-summer for the support of game fish are adequately low in these streams with the possible exception of the lower portions of Sinking Creek and Big Stony Creek. Presence of pools and shelter with adequate food and freedom from predators, including man, appear controlling factors on abundance of fishes in these streams.

Vegetation: Vegetation in the streams which might provide food and shelter, was found predominantly scant at most localities in keeping with the general characteristics of most mountainous streams. The term "scant" for vegetation has been described (Hoover, et al., 1937) as a situation where aquatic vegetation is seldom seen in the streams or the marginal waters, i.e., it does not exceed 10% in area coverage. The term "common" is applied to conditions where 10-50% of exposed rocks are with patches of vegetation or where marginal growth in the water occupies from 10-50% of the stream. Vegetation is extremely important in providing food for organisms which in turn serve as food for fry and fingerlings. It is likely that stream velocity and the type of bottom determines the abundance of vegetation. The mountainous streams examined, with variable flow over

sandy and gravelly bottoms where scouring action is possible, appear unlikely to support an adequate vegetation. The type of bottom found in most of the mountain streams, characteristically of fine to coarse gravel with rubble on sand, is detrimental to the establishment and maintenance of extensive vegetation.

Cover Evaluation: This is an estimation of available shelter and shade afforded a stream from each bank. Shade is a factor which influences temperature conditions in mid-summer, and in many localities an open stream which runs through pasture land and which is susceptible to warming by bright sunlight, particularly at pools, may be subjected to modification and control in this particular through provision of shore-line trees and shrubs. Cover or shade on the stream is estimated by judging how far the cover from each or either bank overhangs the stream. View of this is always taken from the middle of the stream, facing upstream, and values for left bank, right bank, and middle of the stream are assigned on the basis of a scale from 1 to 10, with the larger number indicating most adequate shade. Thus, a reading of $L_0R_0S_0$ would indicate minimum shade if the sun is directly overhead, while $L_{10}R_{10}S_{10}$ would designate a situation where adequate cover conditions permitted shading of the entire stream or pool at the locality with the sun directly above. It is therefore obvious that with but slight density of growth, gain in cover grade can sometimes be accomplished through marked overhang as well as spreading of tree limbs over the stream. Cover evaluation is high for the forested mountain areas mentioned in this report (See Tables I-IV), with clearings and low cover grades assigned to the lowland streams.

Bottom: In the case of these streams, the bottom is composed of broken bedrock, sand, and gravels or rubble of varying degrees of size. Hoover (1937), in the New Hampshire report, states that bottom is of great importance, not only with respect to food production and as a region for attachment of food organisms, but also as suitable fish spawning areas. The types of bottom are productive of food organisms in the following decreasing order: 1. Silt. 2. Small rubble. 3. Large rubble. 4. Coarse gravel. 5. Fine gravel. 6. Bedrock. 7. Sand. Bedrock does not always afford attachment surface suitable for organisms, and shifting sand displaces and smothers organisms. The rubble and coarse gravel of these Giles County streams doubtless have afforded cover and attachment areas. This is especially notable with respect to the numbers of insect larvae representing many Orders.

Chemical Characteristics: The water analyses were carried out at the sites of collection and at the time of collection, following the usual field methods suggested by Hazzard (1935), by Theroux, Eldridge and Mallmann (1936), and by the Standard Methods of the A.P.H.A. (1936), and as previously practiced (Shoup, 1940, 1943, 1944; Shoup, Peyton and Gentry, 1941). Free carbon dioxide (titratable), methyl orange alkalinity, phenolphthalein alkalinity, and dissolved oxygen are all expressed in terms of parts per million (p.p.m.). pH_1 represents the estimation of initial hydrogen-ion concentration, while pH_2 indicates the hydrogen-ion concentration of the sample following complete aeration into equilibrium with atmospheric carbon dioxide. The hydrogen-ion concentration,

estimated in terms of the pH scale, was checked by colorimetric methods, using the Taylor apparatus. In unpolluted natural waters such as these, most chemical factors are the amount of dissolved buffer substances, together with variable quantities of iron, silica, and anions such as chlorides, sulphates and nitrates. Field analyses are usually directed toward estimation of the buffer materials, or the constituents of the bicarbonate buffer system. Lamar and Whetstone (1945-1946), studying the chemical character of Virginia surface waters, have shown the influence of limestone in New River toward contribution to the alkali reserve of this stream, and Shoup (1944) has shown, through results from 371 samplings in Tennessee streams, that frequently the chemical character of an entire stream can be correlated with the solubility of buffers in its predominant bedrock formation. In general, streams draining from siliceous formations such as the Pre-Cambrian and Cambrian of East Tennessee or the Clinch Sandstone, will carry waters that are decidedly soft and are frequently mentioned as being the "free-stone" streams by natives of mountainous areas. The quantity of total buffer as bicarbonate in such streams will not be sufficient to offset change in pH by small amounts of dissolved carbon dioxide, so that characteristically such streams will have pH values on the acid side of neutrality because of predominance of carbonic acid over the bicarbonate present. The amount of carbon dioxide in the atmosphere in equilibrium with the water is quite enough to bring the pH to 6.0. Such conditions are characteristic of Little Stony Creek, Pond Drain, Hunter's Branch, and the upper part of Big Stony Creek where the Clinch Sandstone is the dominant surface formation and layers of dolomite are absent. In such areas, standing waters of bogs or swampy spots drop to very low pH values where both carbon dioxide and humic acids may be derived from vegetation. Near Station 5 alongside Little Stony Creek, and opposite the residence estate of White Pine Lodge may be found a bog embedded deep in thicket. This, at an altitude of 3174 ft. lies in a region free of limestone. Samples of the bog waters, taken from a mat of the vegetation, gave free carbon dioxide values of 63.0 p.p.m.; M.O. alkalinity of 8.0 p.p.m., and oxygen concentration of but 4.0 p.p.m. The initial samples had pH₁ values of 4.8, which on being aerated to eliminate volatile acid (CO₂), gave pH₂ of 5.2.

In the lower portions of these streams (Little Stony Creek and Big Stony Creek) and in those located in the dolomite formations (Spruce Run and Sinking Creek), soluble carbonate as both magnesium and calcium carbonates slowly changes the character of the waters to bring the pH values to 8.0 or above, with a rapid rise to more than 100 parts per million of total alkalinity as determined by titration. Excess carbonate, which is not held as bicarbonate (HCO₃) in the presence of carbon dioxide, is demonstrated by titration with phenolphthalein as indicator, and is referred to as the "phenolphthalein alkalinity" (P. Alk.), and this consequently means no free carbon dioxide is available in such waters excepting in pools. Spruce Run is the only stream of the group bearing these characteristics, combining a carbonate (CO₃) alkalinity with no free carbon dioxide and a quite high total alkalinity (122.0-135.0 p.p.m.) characteristic of a hard-water stream. Aquatic areas of this kind are typical lowland habitats of a good degree

of productivity in terms of bottom organisms and "wet food" volume. Headwaters of New River (Lamar and Whetstone, 1945-1946) are reported to be fairly

TABLE I

Limnological Data of Sinking Creek, Craig and Giles Counties, Va.

	STATION 1	STATION 2	STATION 3
Date	July 28, 1947	July 28, 1947	July 29, 1947
Air T., °F.	75.5°	76.0°	83.0°
Water T., °F.....	70.5°	69.0°	80.0°
Water T., °C.	21.3°	20.5°	27.4°
Hour.....	3:00 P.M.	4:10 P.M.	3:30 P.M.
Altitude.....	2200 Ft.	1800 Ft.	2300 Ft.
Av. Width.....	45 Ft.	25 Ft.	22 Ft.
Av. Depth.....	6 In.	2 Ft.	10 In.
Vol. Flow	20.0 Cu. Ft. Sec.	25.0 Cu. Ft. Sec.	6.0 Cu. Ft. Sec.
Velocity	1.1 Ft. per Sec.	0.5 Ft. per Sec.	0.2 Ft. per Sec.
Turbidity.....	Clear	Clear	Clear
pH ₁	8.0	7.9	7.9
pH ₂	8.1	8.1	8.1
Free CO ₂	9.0 p.p.m.	8.0 p.p.m.	7.0 p.p.m.
M.O. alkalinity.....	110.0 p.p.m.	122.5 p.p.m.	118.0 p.p.m.
P. alkalinity.....	Zero	Zero	Zero
Oxygen.....	9.2 p.p.m.	9.2 p.p.m.	8.2 p.p.m.
Bottom.....	Sand. Fine gravel	Sand. Fine and coarse gravel	Sand. Fine gravel
Vegetation.....	Scant	Scant	Scant
Cover Grade.....	L ₃ R ₇ S ₀	L ₃ R ₃ S ₀	L ₃ R ₂ S ₀
Food available:			
Annelida.....	—	—	+
Mollusca.....	+	+	+
Arachnida.....	+	+	+
Crustacea.....	+	+	+
Plecoptera.....	+	+	—
Ephemeraida.....	+	+	+
Odonata.....	—	—	+
Hemiptera.....	—	+	—
Tricoptera.....	+	+	—
Diptera.....	—	+	+
Coleoptera.....	+	—	+
Neuroptera.....	—	—	—
Predators.....	—	—	—
Food volume.....	4.0 cc.	3.0 cc.	3.0 cc.
Food Grade.....	1	1	1

low in total alkalinity and gain in bicarbonate along the watercourse. Lamar and Whetstone (1945-1946) report M.O. alkalinity values for New River at Eggleston as 50-52 p.p.m. and pH 7.3-7.4. We have examined New River at the highway bridge 1 mile below Pearisburg, at a point where limestone washes from quarries enter the river, and have found the following characteristics:

New River: 1 mile below Pearisburg, Va.

July 31, 1947.

Air T°.: 89.0°F. Water T°.: 80.0°F.

pH₁ = 8.0 pH₂ = 8.0

Methyl Orange Alk. = 50.0 p.p.m.

Phenolphthalein Alk. = 2.0 p.p.m.

Dissolved Oxygen = 8.0 p.p.m.

TABLE II

Limnological Data of Spruce Run Creek, Giles County, Va.

	STATION 6	STATION 7
Date..	July 31, 1947	July 31, 1947
Air T., °F.....	81.0°	79.0°
Water T., °F.....	65.0°	67.0°
Water T., °C.....	18.3°	19.5°
Hour.....	9:50 A.M.	10:30 A.M.
Altitude	2027 Ft.	1662 Ft.
Av. Width.....	7.0 Ft.	12.0 Ft.
Av. Depth.....	3.0 In.	4.0 In.
Vol. Flow.....	3.0 Cu. Ft. Sec.	8.0 Cu. Ft. Sec.
Velocity	1.0 Ft. Sec.	1.0 Ft. Sec.
Turbidity.....	Clear	Clear
pH ₁	7.9	8.2
pH ₂	7.9	8.2
Free CO ₂	Zero	Zero
M.O. alkalinity.....	122.0 p.p.m.	135.0 p.p.m.
P. alkalinity.....	3.7 p.p.m.	7.3 p.p.m.
Oxygen.....	8.8 p.p.m.	10.0 p.p.m.
Bottom.....	Sand. Fine gravel	Sand. Coarse gravel
Vegetation	Scant. Some algae	Scant. Sparse algae
Cover Grade.....	L ₃ R ₃ S ₀	L ₇ R ₁ S ₀
Food available:		
Annelida.....	—	—
Mollusca.....	+	+
Arachnida.....	—	—
Crustacea.....	+	+
Plecoptera.....	+	+
Ephemera.....	+	+
Odonata.....	+	—
Hemiptera.....	—	—
Tricoptera.....	+	+
Diptera.....	—	—
Coleoptera.....	—	—
Neuroptera.....	—	—
Predators.....	Turtles. Snakes	Snakes abundant
Food volume.....	5.0 cc.	3.0 cc.
Food Grade.....	1	1

Natural waters of these qualities, which are unpolluted, will generally carry adequate quantities of dissolved oxygen in parts per million for support of fishes

and other fauna. Embury (1927) has indicated that brook trout waters should contain not less than 3 cc. of oxygen per liter, or 4.29 parts per million. Rocky mountainous streams, well-aerated through their fall over rubble and boulders

TABLE III
Limnological Data of Little Stony Creek, Giles County, Va.

	STATION 4	STATION 5	STATION 8
Date.....	July 30, 1947	July 30, 1947	July 31, 1947
Air T., °F.....	72.0°	81.0°	84.0°
Water T., °F.....	55.0°	61.0°	68.0°
Water T., °C.....	12.8°	16.0°	19.9°
Hour.....	10:30 A.M.	3:00 P.M.	11:30 A.M.
Altitude.....	3553 Ft.	3174 Ft.	1690 Ft.
Av. Width.....	6.0 Ft.	10.0 Ft.	15.0 Ft.
Av. Depth.....	3.0 In.	4.5 In.	6.0 In.
Vol. Flow.....	10.0 Cu. Ft. Sec.	20.0 Cu. Ft. Sec.	30.0 Cu. Ft. Sec.
Velocity.....	0.66 Ft. Sec.	0.83 Ft. Sec.	0.50 Ft. Sec.
Turbidity.....	Clear	Clear	Clear
pH ₁	6.0	6.2	7.0
pH ₂	6.0	6.1	7.2
Free CO ₂	3.3 p.p.m.	2.0 p.p.m.	2.5 p.p.m.
M.O. alkalinity.....	4.0 p.p.m.	6.0 p.p.m.	17.0 p.p.m.
P. alkalinity.....	Zero	Zero	Zero
Oxygen.....	9.2 p.p.m.	8.04 p.p.m.	9.20 p.p.m.
Bottom.....	Sand. Fine gravel. Bedrock	Sand. Fine gravel. Boulders	Sand. Fine and coarse gravel
Vegetation.....	Common	Abundant	Scant
Cover Grade.....	L ₁₀ R ₁₀ S ₅	L ₁₀ R ₁₀ S ₁₀	L ₅ R ₇ S ₀
Food available:			
Annelida.....	—	—	—
Mollusca.....	—	—	—
Arachnida.....	+	—	—
Crustacea.....	+	+	+
Plecoptera.....	+	+	+
Ephemera.....	+	+	+
Odonata.....	—	—	—
Hemiptera.....	—	—	—
Tricoptera.....	+	+	+
Diptera.....	—	—	+
Coleoptera.....	—	—	+
Neuroptera.....	—	+	—
Predators.....	—	—	—
Food volume.....	—1.0 cc.	2.0 cc.	1.0 cc.
Food Grade.....	3	2	2

and through riffles, and free of pollution, may always be counted on as having a sufficient quantity of dissolved oxygen; an amount which is dependent upon the temperature of the waters. In some lowland streams, in warmer waters, and in pools, the oxygen saturation value may fall, but unless pollution is present, a

dangerously-low level for support of aquatic organisms is not likely to be attained. These streams of the east bank of New River all demonstrate an oxygen concentration in step with the water temperatures.

TABLE IV
*Limnological Data of lower Big Stony Creek, Pond Drain, and Hunter's Branch,
Giles County, Va.*

	BIG STONY CREEK STATION 9	POND DRAIN STATION 10	HUNTER'S BRANCH STATION 11
Date.....	July 31, 1947	August 1, 1947	August 2, 1947
Air T., °F.....	89.0°	75.0°	70.0°
Water T., °F.....	75.0°	63.0°	55.0°
Water T., °C.....	23.8°	17.2°	12.8°
Hour.....	3:20 P.M.	4:30 P.M.	11:20 A.M.
Altitude.....	2100 Ft.	3790 Ft.	3821 Ft.
Av. Width.....	10.0 Ft.	2.5 Ft.	0.5 Ft.
Av. Depth.....	6.0 In.	2.0 In.	1.0 In.
Vol. Flow.....	35.0 Cu. Ft. Sec.	1.0 Cu. Ft. Sec.	0.33 Cu. Ft. Sec.
Velocity.....	1.0 Ft. Sec.	1.0 Ft. Sec.	0.7 Ft. Sec.
Turbidity.....	Clear	Clear	Clear
pH ₁	7.1	6.2	6.3
pH ₂	7.2	6.4	6.4
Free CO ₂	1.0 p.p.m.	7.0 p.p.m.	2.0 p.p.m.
M.O. alkalinity.....	23.0 p.p.m.	6.0 p.p.m.	5.0 p.p.m.
P. alkalinity.....	Zero	Zero	Zero
Oxygen.....	8.0 p.p.m.	9.20 p.p.m.	9.70 p.p.m.
Bottom.....	Sand. Fine and coarse gravel Boulders	Sand. Fine and coarse gravel.	Sand. Fine and coarse gravel
Vegetation.....	Scant	Scant	Scant. Some mosses
Cover Grade.....	L ₆ R ₄ S ₀	L ₁₀ R ₁₀ S ₁₀	L ₁₀ R ₁₀ S ₁₀
Food available:			
Annelida.....	—	—	—
Mollusca.....	—	—	—
Arachnida.....	—	—	+
Crustacea.....	+	+	+
Plecoptera.....	+	—	+
Ephemera.....	+	+	+
Odonata.....	—	—	—
Hemiptera.....	—	+	+
Tricoptera.....	+	+	+
Diptera.....	+	+	—
Coleoptera.....	—	—	—
Neuroptera.....	+	—	—
Predators.....	—	—	—
Food volume.....	2.0 cc.	0.2 cc.	3.0 cc.
Food Grade.....	2	3	1

The factors mentioned above as isolated features of the environment have relatively little influence on the productivity of food in a stream and the presence of large numbers or of many species of fishes. However, in the aggregate, tem-

peratures that are high enough to promote rather than retard growth, dissolved substances providing buffer action and mineral nutrition, an adequate oxygen supply, coupled with suitable cover and shelter, all do tend to support increase in numbers and species of food organisms. In absence of predators, the numbers and distribution of fishes should be beneficially influenced by the sum of these conditions. It is worthy of note that Burton and Odum (1945) found a rather wide distribution of fishes in both Sinking Creek and Spruce Run Creek and a somewhat more restricted distribution in terms of numbers of species in Little Stony Creek. If it is true, as Needham (1938) indicates, that food for game fishes is more abundant in the rather alkaline waters, our observed values for food volume and food grade would indicate production of more fish flesh per unit area in Sinking Creek and Spruce Run.

SUMMARY

1. Certain limnological characteristics of streams draining into the east bank of New River in the vicinity of Mountain Lake, Virginia, have been determined as a part of the work in limnology at the Mountain Lake Biological Station in the summer of 1947.

2. Field determinations of general water chemistry and productivity show these streams to be unpolluted and with chemical and thermal characteristics related to altitude and to the geological formations comprising the dominant bedrock.

3. In general, chemical and productivity features of Pond Drain, Hunter's Branch, Little Stony Creek, and the upper half of Big Stony Creek are all characteristic of mountain streams flowing through siliceous strata free of substances contributing to the alkali reserve. In contrast, within a short distance of the above, Sinking Creek and Spruce Run Creek show the characters of lowland alkaline streams of somewhat greater productivity potential in direct relation to the dominant dolomite strata comprising the bedrock of these streams.

4. The information obtained is a contribution toward knowledge of the biogeochemical relationships in fresh waters.

LITERATURE CITED

- AMERICAN PUBLIC HEALTH ASSOCIATION. 1936. Standard Methods for the examination of water and sewage. 8th ed. pp. 1-309. N. Y.
- BURTON, G. W., AND E. P. ODUM. 1945. The distribution of stream fish in the vicinity of Mountain Lake, Virginia. *Ecology* **26** (2): 182-194.
- BUTTS, CHARLES. 1933. Geologic Map of the Appalachian Valley of Virginia with explanatory text. Vir. Geol. Survey Bull. No. 42, pp. 1-56, with map. University, Va.
- EMBODY, G. E. 1927. An outline of stream study and the development of a stocking policy. Contrib. Aquicultural Lab., Cornell Univ., pp. 1-21. Ithaca, N. Y.
- HAZZARD, A. S. 1935. Instructions for stream and lake survey work. U. S. Bur. Fisheries. (mimeo.) pp. 1-34. Washington.
- HOOVER, E. E., ET AL. 1937. Biological Survey of the Androscoggin, Saco and Coastal Watersheds. Methods of stream and lake analysis. Survey Rept. No. 2, pp. 128-150. New Hampshire Fish and Game Dept., Concord.
- LAMAR, W. L., AND G. W. WHETSTONE. 1947. Chemical character of surface waters of

- Virginia. (1945-1946). Vir. Conservation Comm., Div. of Water Resources and Power, Bull. No. 8, pp. 1-46. Charlottesville, Va.
- NEEDHAM, P. R. 1938. Trout Streams: Conditions that determine their productivity and suggestions for stream and lake management. pp. 1-233. Comstock Publ. Co., Ithaca, N. Y.
- SHOUP, C. S. 1940. Biological and chemical characteristics of the drainage of the Big South Fork of the Cumberland River in Tennessee. Tenn. Div. of Game and Fish, Misc. Publ. No. 1, pp. 76-105. Nashville.
- 1943. Distribution of fresh-water gastropods in relation to total alkalinity of streams. *Nautilus* 56 (4): 130-134.
- 1944. Geochemical interpretation of water analyses from Tennessee streams. *Trans. Amer. Fisheries Soc.* 74: 223-229. (Publ. 1947).
- , J. H. PEYTON, AND GLENN GENTRY. 1941. A limited biological survey of the Obey River and adjacent streams in Tennessee. Tenn. Div. of Game and Fish, Misc. Publ. No. 3, pp. 48-76. Nashville.
- THEROUX, F. R., E. F. ELDRIDGE, AND W. LER. MALLMANN. 1936. Laboratory manual for chemical and bacterial analysis of water and sewage. 2nd ed. pp. 1-228. N. Y.

A COMPARATIVE STUDY OF ZYGOTE GERMINATION IN THE SAPROLEGNIACEAE*

BY ARTHUR WILLIAM ZIEGLER

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina*

PLATES 1-6

The Saprolegniaceae have been of interest to botanists since 1823 when Nees von Esenbeck founded the genera *Saprolegnia* and *Achlya*. Scores of papers have now been written on the members of this family by men too numerous to mention here. These papers have covered morphology, taxonomy, cytology, and physiology of the species of this family so thoroughly that today it is perhaps the best known of the families of the Phycomycetes. Coker and Barnhart (1937) and Wolf (1944) give rather complete bibliographies of this group of fungi.

In spite of the great amount of work that has been done on these fungi, we are still lacking information on the complete life history of a number of the Saprolegniaceae. Most accounts of the species development of these forms stop at the fertilized egg which usually then undergoes a rest period before germination. The germinating oospore has been seen only in relatively few cases. This lack of information has probably been due to the varied length of rest of these species, and either to a supposed difficulty in germinating the eggs or a lack of interest in the procedure.

It was felt, therefore, that a worthwhile contribution could be made to this field by attempting to germinate the resting zygotes of several species, by performing several physiological experiments on the resting oospores, and by gathering together the literature on this subject which is spread over a period from 1860 to 1937.

HISTORICAL NOTES

In the literature of this family several terms are used synonymously. For example, the words oospore, egg, and zygote are used to connote the fertilized structures contained within the oogonium. While it is felt that the word zygote more definitely implies the fertilized egg, the other terms, through custom and long usage are retained in this paper.

In 1860, Anton de Bary published his description of a new genus, *Aphanomyces*, with several species. He observed the germination process in *A. stellatus* which he had maintained in a water culture for three months. De Bary described the gradual darkening of the oospore with its granular protoplasm and the putting forth from the oospore of a cylindrical germ tube which pushed through the

* This paper was condensed from a dissertation presented to the faculty of the University of North Carolina in partial fulfillment of the requirements for the degree of Doctor of Philosophy in the Department of Botany.

oogonial wall to the outside. This tube branched immediately several times and the cytoplasm flowed into it until the oospore proper became empty.

Pringsheim (1873-1874) recorded the germination of the oospores of *S. ferax* by a short or long germ tube with a sporangium produced at the end, or a branched mycelium with several sporangia. In *A. polyandra* he showed the changes in the oospore from the mature egg to the short germ tube with a sporangium. Finally, he showed the short or long germ tube formation with probable zoosporangia in *A. racemosa*.

Sorokine (1876) practically repeated de Bary's work on *Aphanomyces*. He found that the oospores would germinate in the manner described by de Bary after three months of rest, or would germinate by producing a short germ tube which broke out of the oogonial wall and formed zoospores within 24 hours. Germination occurred only if the oospores remained in the light.

De Bary also mentioned zygote germination in his book on the fungi, mycetozoa, and bacteria. In a new species, *Aplanes Braunii*, he referred to the formation of a new thallus directly from the germ tube which developed from the germinating oospore.

While working on the karyology of *Saprolegnia*, Trow, in 1895, saw the germinating oospores of *S. Thureti*. He noted that the oospore wall became thin, the oil globule disappeared, and a germ tube was produced which formed a club-shaped sporangium.

In 1899, Trow, working on *Achlya americana* var. *cambrica*, gave a complete account of the germinating oospore. He pointed out that within six days after the oospore had reached maturity the inner wall began to dissolve, and the protoplasm increased in quantity. Meanwhile, the oil globule moved to center and was gradually decomposed and absorbed. The outer wall stretched, the inner wall completely disappeared, and the oospore increased in size with a vacuole in the center. After this, one or more germ tubes made their appearance, ceased growth, and produced small sporangia with four to ten zoospores. Trow also tried to hasten germination by subjecting the oospores to high temperature and ice but to no avail.

Klebs (1899) found that the oospores of *S. Thureti* (*S. mixta*) would either germinate by a short germ tube with an apical sporangium, or would form a mycelium directly on germination.

In 1911, Ksanowsky found the germinating oospores of *Aphanomyces laevis*. Germination had not been seen in this species prior to this time probably because the resting period of this form lasted about seven months. His findings, however, closely parallel those of de Bary for *A. stellatus*.

Not until 1918 was another record of oospore germination made. Weston, working on the development of *Thraustotheca clavata*, found that mature oospores placed in pure spring water maintained at 25° C. would germinate. Under these conditions the protoplasmic content increased, and the oil droplet became irregular and gradually disappeared. Meanwhile, the wall first swelled and was resorbed. It then became distended until it was comparatively thin. A hypha

was put out from the zygote which either formed a sporangium or grew into an extensive mycelium if enough nutrient were present.

In his book, "The Saprolegniaceae" (1923), Coker described the germinated eggs of *Achlya apiculata* var. *prolifera*. After 48 hours on agar the eggs sprouted to form long, branched threads.

The following year (1924), Coker and Couch, working with *Thraustotheca primoachlya*, found that the oospores germinated in the manner of those of *T. clavata* described by Weston. At room temperature the oospores would sprout in from ten days to two weeks.

Jones and Drechsler, in 1925, working on *Aphanomyces euteiches*, found that this form would germinate after a fifteen-day rest. Material from agar cultures was transferred to hanging drops and the oospores germinated either by mycelial development or by production of zoospores. The type of germination depended on the amount of nutritive material present. Mycelial growth occurred whenever particles of substratum were allowed to remain in the culture, and zoospore production when the material was allowed to soak first. The changes in the oospore prior to germ tube formation closely resemble those described by Weston for *T. clavata*.

While working on the problem of heterothallism in *Dictyuchus*, Couch (1926) was able to induce oospore germination after a resting period of six weeks to two months by placing the ripe oospores in boiled corn-grain juice or, as he later found, in boiled well water. The oospore changes before germination parallel those described above.

Patterson (1927) showed the beginning of the germ tube with nuclei and cytoplasm in *Achlya colorata*.

Schlösser (1929), working on several species of *Saprolegnia* whose normal resting periods were from three weeks to three months, was able to reduce this period somewhat by freezing the eggs for three days. He also found that germ tubes of individual zygotes do not always make their way through the top of the oogonium but mostly push through the oogone membrane at any convenient place.

In some unpublished observations on zygote germination in *Achlya orion* and *A. flagellata*, Couch (1933) found that the zygotes of *A. flagellata* were transformed into sporangia which opened by small tubes to the outside, allowing the eight or ten zoospores to escape. In *A. orion* he observed the changes prior to germination and found that these would germinate by rather long germ tubes with a sporangium formed at the end.

Latham, in 1935, refound Cornu's *Achlya recurva*. In this species the oospores mature in three days and are capable of immediate germination. Changes in the structure of the zygote prior to germ tube formation were not described. There was usually a single germ tube from each egg which grew out to a distance of 50-75 micra with a sporangium formed at the tip.

In 1937-1938 Schrader worked on the development of *T. clavata* and found that the oospores of this species have a rest period from twenty days to a much

longer time. He showed the two divisions of reduction division in the oospore, several germinational changes in the oospore, and the formation of the germ tube. He did not see the formation of sporangia, as they would not develop in his hanging drop preparations.

MATERIALS AND METHODS

Extensive collections of soil and water were made by the author in eastern and central North Carolina and northern and eastern South Carolina. The material was brought to the laboratory and baited with boiled split hemp seeds. Tap water mixed with charcoal, filtered, and autoclaved, was used for the soil cultures and rough isolation. Pure cultures of each form were made in the usual way on maltose-peptone agar #5. All final determinations of species were made from cultures grown on hemp seeds using distilled water which had been treated with charcoal, filtered, and autoclaved.

While awaiting the end of the resting period of the eggs, a pure culture of each form studied was placed in a 250-cc. Erlenmeyer flask with 150 cc. of charcoal-filtered distilled water. Several split hemp seeds served as food. The flasks were plugged with cotton and left on the laboratory table. This has proved to be an excellent way to keep cultures alive for several months and to avoid weekly washing and adding of food.

To observe the germinating eggs easily a small portion of the fungus mycelium was removed from the flask and placed in fresh charcoal distilled water on a slide. The water drop was spread out so the lens of the high objective would just avoid being wet. The whole slide was placed in a petri dish and additional water was added to prevent drying. The zygotes were easily seen under high dry power using this method, and the mycelium usually stayed in place on the slide when the stale water was removed and fresh added each morning by careful pipetting.

Germination studies were begun in an upstairs laboratory until a period of hot weather apparently stopped the germinating zygotes. The studies were then made in a basement where the temperature range during the entire study varied from 72° to 82° F. Temperatures above 85° F. seem to inhibit egg germination.

Additional techniques involving treatments other than those described above are written up under each experiment performed.

EGG GERMINATIONS

Aplanes treleaseanus (Humph.) Coker

Plate 1, figures 1-9

Aplanes treleaseanus was isolated from a soil collection and was seven weeks and one day old when placed in fresh water at 11:30 A.M. on June 25, 1947 (Fig. 1). The temperature during its development ranged from 76° to 81° F.

This specimen of *A. treleaseanus* differs from Coker's (1923) description in that the egg wall is not thick but relatively thin. However, a dark line of cytoplasm was noted immediately inside the egg wall, which might indicate that the eggs

had begun to germinate but ceased their development due to accumulation of staling products. This thin wall, then, might be due to this factor. Germination continued in fresh water and at 2:00 P.M., June 25, some of the cytoplasm had lightened somewhat but several dark areas had appeared in the periphery of the cytoplasm adjoining the oil droplets (Fig. 2). Eggs that are to disintegrate also had this same appearance rendering it difficult to determine whether an egg was to disintegrate or germinate. At 7:30 P.M. the oil droplets had become scattered throughout the cytoplasm and the dark area immediately beneath the wall had disappeared (Fig. 3). There was a slight increase in egg size and the wall was definitely much thinner. By 8:05 P.M. the cytoplasm had become less dense and a small vacuole had appeared (Fig. 4). At 9:30 P.M. the vacuole had increased in size and a small protrusion had formed on the side of the egg. A slight increase in egg size was noted (Fig. 5). By 1:00 A.M., June 26, the germ tube had elongated and passed to the outside of the oogonium through an oogonial pit. The cytoplasm was light in color with small glistening oil droplets scattered throughout (Fig. 6). The germ tube was observed at 9:00 A.M. and at 12:00 M. on June 26, but only an increase in length was noted. By 2:00 P.M. a cross wall had delimited the sporangium, which was filled with lightly colored, vacuolate cytoplasm (Fig. 7). This sporangium was not observed until 7:45 P.M. when nine spores had been formed (Fig. 8). By 9:30 A.M., June 27, each spore had begun to sprout.

The germ tube shown in detail in the drawings is not typical of most of those seen in the culture. Most of these were very long tubes as can be seen by the drawing made at 178 magnification (Fig. 9).

The stages in the germination of the eggs of *A. treleaseanus* resemble closely those shown and described by de Bary (1884) for *A. braunii* (*A. androgynus*), although he did not describe any detailed steps before germ tube appearance. In this species, de Bary pictures the germ tube as being short with a terminal sporangium. Short tubes do occur in *A. treleaseanus* but the germ tube usually is quite long before a sporangium is formed.

Germination began immediately after a portion of the mycelium was placed in fresh water and was completed within thirty-two hours.

Saprolegnia ferax (Gruith.) Thuret

Plate 1, figures 10-16

The culture of this form was five weeks and two days old when placed in fresh water on June 24, 1947, at 11:30 A.M. (Fig. 10).

The first stage in germination—a splitting up of the oil droplets and a darkening of the cytoplasm—was seen at 2:20 P.M., June 28, 1947 (Fig. 11). By 7:15 P.M. the same day, the oil particles had scattered throughout the cytoplasm and the egg wall had a scalloped appearance on the inner side. A definite increase in size was noted (Fig. 12). At 10:00 A.M., June 29, the egg wall had thinned considerably and a central vacuole had developed. A small germ tube had begun to protrude from the egg (Fig. 13). This tube had passed through

an oogonial pit to the outside by 4:00 P.M. (Fig. 14). After twenty-two hours (2:30 P.M., June 30) the germ tube had grown in length somewhat and the egg cytoplasm had passed out into the tube (Fig. 15). Another egg in the oogonium, meanwhile, had germinated in the manner described above. The germ tubes were rather short and thick and cross walls formed in the germ tubes delimiting the sporangia near their exit from the oogonial pits. Seven spores were liberated from the sporangium of the first germinating egg at 8:30 P.M., June 30, 1947 and the second germ tube sporangium later released five spores.

Germination began three days and twenty-two hours after the mycelium was placed in fresh water and was completed within two days and seven hours after its inception.

Saprolegnia delica Coker

Plate 2, figure 4

Saprolegnia delica was eight weeks and two days old when it was placed in fresh water at 3:00 P.M., July 7, 1947. During its development the temperature ranged from 72° to 77° F. The first evidence of germination was noticed seven days and one hour after the mycelium was placed in fresh water and was completed within three days and twelve hours after it began. The stages of germination closely resemble those described for *S. ferax*. The drawing shows an oogonium in which four eggs have germinated.

Saprolegnia megasperma Coker

Plate 2, figures 1-2

The culture of this form was twelve weeks and two days old when placed in fresh water. The germination of this large species had been attempted six other times before this but with no success.

Early germination stages resemble those of other members of the genus *Saprolegnia* with no variation from the usual pattern. A large number of germinated zygotes were seen. In the majority of cases, the germ tube ceased growth very shortly and produced a single short sporangium (Fig. 1). In other cases a very long germ tube developed with a small sporangium at the apex. Two instances of a germ tube forming several functional sporangia were observed. One of these is shown in Figure 2. In all cases, the spores which were liberated behaved as in *Saprolegnia*.

Germination began thirty hours after the mycelium was placed in fresh water. The process was completed two days and twenty-two hours after it began.

Saprolegnia dichina Humphrey

Plate 2, figures 5-6

This species, collected at Chapel Hill, N. C., was two months and five days old when placed in fresh water at 10:00 A.M. on October 15, 1947.

The first evidence of germination—a breaking up of the oil globules—was seen

at 2:20 P.M. on October 16. The germination of this species so closely resembles *S. ferax* and the other species of *Saprolegnia* that one is referred to those species for the details of the process.

By 8:35 A.M., October 18, a long germ tube had been formed by the oospore and an apical sporangium was being formed (Fig. 2). This discharged about eight spores at 11:50 A.M. the same day (Fig. 6). Meanwhile, a second egg in the same oogonium had germinated but a very short germ tube was formed with a rather clavate sporangium. The more usual type is the former one and these were fairly abundant in my culture (Fig. 5).

The first stage in germination was seen twenty-eight hours after the mycelium was placed in fresh water and was completed within an additional forty-five hours.

Saprolegnia litoralis Coker

Plate 2, figure 3

Saprolegnia litoralis had remained in the flask for eleven weeks and one day before being placed in fresh water at 2:30 P.M., July 24, 1947.

The beginning of development was not observed until 9:05 A.M. on August 2. Its early development resembled both *S. ferax* and *S. delica* and the early stages described for those forms are applicable to this species.

The germ tubes were quite large and usually fairly short. They passed to the outside through the large and conspicuous oogonial pits. Cross walls formed either long-cylindrical or slightly bulbous sporangia. Examples of both are shown—the two types occurring from germinating eggs in the same oogonium (Fig. 3). The sporangia emptied their spores in the manner typical of *Saprolegnia*.

The first sign of germination was seen eight days and nineteen hours after a portion of the mycelium was placed in fresh water. The process was concluded within two days and five hours after it began.

Isoachlya unispora Coker and Couch

Plate 2, figures 7–11; Plate 3, figure 1

Frequent attempts to germinate the zygotes of this species were unsuccessful until October 1, 1947, when the culture was four months and two days old. The use of the term zygote is debatable here as no antheridia are formed, but it is used merely for convenience.

Previous unsuccessful attempts to germinate this species, *I. intermedia*, and *I. itoana* had included the use of various nutrient decoctions, agar, alkaline and acid solutions, and growth in the incubator and cold room.

A portion of the mycelium was placed in fresh water at 10:20 A.M., October 1, 1947, when the culture was four months and two days old (Plate 2, fig. 7). The first indication of germination was seen at 8:15 A.M., October 3, when the central cytoplasm had darkened and the oil globules appeared to be breaking up into smaller particles (Fig. 8). By 12:20 P.M. the oil layers as such had dis

appeared and glistening particles appeared throughout the darkened cytoplasm. The inner zygote wall was scalloped and ragged in appearance (Fig. 9). During the next twelve hours, the zygote wall had become uniformly thin, the cytoplasm had become less granular, and a central vacuole had formed (Fig. 10).

The egg put out a germ tube which passed through the single conspicuous pit to the outside. This had attained a length equal to the zygote width by 10:50 A.M., October 5 (Fig. 11). It continued to grow during the remainder of the day and most of October 6 and produced sparse branches. A sporangium was formed on the apical end, seven spores were cut out, and these were discharged at 3:10 P.M., October 6, swimming away as in *Saprolegnia*. Several other sporangia were formed in a cymose arrangement as seen in Plate 3, figure 1.

In my culture, a large number of zygotes germinated. Eighty per cent germinated in the manner described above. The remainder formed a long, branched, intertwining, hyphal system.

Forty-six hours after the mycelium was placed in fresh water the first sign of germination was seen. The germination process required three days and seven hours.

Isoachlya intermedia (Coker and Harv.) Coker

Plate 3, figure 2

The culture of this form was five months and three weeks old when the zygotes germinated. Many previous attempts to germinate this species had failed. (See *I. unisporea*.) The zygotes would germinate only when they were placed in a weak corn-grain solution (Couch, 1926). The zygotes could not be germinated in fresh water.

The details of the germination process resemble those described for *I. unisporea*. In the zygote followed through its development, only a much branched, long mycelium was formed. In other cases, the germinating zygote produces not only a long branched mycelium but also zoosporangia. This last type may be seen in Figure 2.

Germination began twenty-three hours after the culture was placed in corn-grain juice. The process was complete about thirty-one hours after it began.

Isoachlya itoana Nagai

Isoachlya itoana Nagai was collected by Mr. Arthur Kelman of State College, Raleigh, N. C., who identified it and kindly gave me a culture.

Several attempts to germinate the zygotes of this species had resulted in failure. Germination was obtained, however, by placing a culture five months and one week old in corn-grain juice. (See *I. intermedia*.) A second culture was placed in fresh water but failed to germinate.

Details of the germination process are like those figured and described for *I. unisporea* and *I. intermedia*. The germ tube in this species finally formed a much branched mycelium with a single sporangium. Only five zygotes have been seen to germinate and all formed a mycelium and a single zoosporangium.

Germination began twenty-six hours after the mycelium was placed in the

corn-grain decoction. Spores were liberated two days and fourteen hours after germination.

Achlya colorata Pringsheim

Plate 3, figures 3-8; Plate 4, figure 1

My culture of this fungus was five weeks and four days old when placed in fresh water on July 7, 1947, at 2:30 P.M. (Plate 3, fig. 3).

* One egg had begun germinating by 8:20 A.M., July 8. The cytoplasm had darkened noticeably beneath the wall and the two layers of oil droplets appeared to be splitting up into smaller fragments. These gradually invaded the clear space immediately beneath the wall. Several dark areas were noticed in the central cytoplasm (Fig. 4). By 2:20 P.M. the scalloped egg wall had become thinner and a slight increase in size of the egg was evident (Fig. 5). At 8:00 P.M. the wall had reached its final thinness and a central vacuole had developed with a slight lightening of the cytoplasm (Fig. 6). A small germ tube had put out from the egg by 9:30 A.M., July 9 (Fig. 7). The only other change noticed during the remainder of the day and July 10 and 11 was an increase in length of the germ tube (Fig. 8). At 4:30 P.M., July 11, the germ tube had attained its maximum length and a cross wall had formed delimiting the sporangium. Spore initials were being formed as the drawing was made (Plate 4, fig. 1). Approximately twenty-two spores were formed and were liberated by 9:15 A.M., July 12.

No mention was made above of the germ tube's passing to the outside of the oogonium. *Achlya colorata* has rather thick oogonial walls with short blunt outgrowths. The germ tube described above apparently passed through the oogonial wall at a thin place in the wall caused by the antheridial tube. A slight constriction of the tube as it passed through this thin spot may be noted on Plate 4, figure 1. However, in several cases, the germ tube in its passage to the outside actually split the oogonial wall. Several examples of this have been seen.

Patterson (1927) had seen the germinating eggs of this form, but there has been no general account of the germination process in this species up to now.

The first evidence of germination was seen twenty hours after beginning the experiment and germination was completed in four days.

Achlya racemosa Hildebrand

Plate 4, figure 2

This species, which had been isolated from a soil collection, was ten weeks and six days old when placed in fresh water at 9:45 A.M., July 27, 1947.

In its development this species parallels closely its related form *A. colorata*. The developing germ tube, however, emerges through a pit caused by an antheridial tube and produces a sporangium typical of *Achlya* (Fig. 2). Numerous discharged sporangia were to be found in the culture with the number of discharged zoospores averaging about eleven.

Forty-eight hours elapsed before the first indication of germination was seen and the process was concluded three days and ten hours after it began.

Achlya flagellata Coker

Plate 4, figures 3-10

My culture of this form was four weeks old when it was placed in pure fresh water at 9:30 A.M. on June 1, 1947. Several germinating eggs were noted on June 4, so a single oogonium was then kept under observation until germination was complete (Fig. 3).

The first evidence of germination was noted at 9:15 A.M., June 5, when the egg had increased very slightly in size and the protoplasm had become darker. The oil droplet moved from the periphery toward the center and became completely surrounded by cytoplasm. Except in the area adjacent to the former position of the oil drop, a general thinning of the wall occurred (Fig. 4). By 2:05 P.M. the same day, a further increase in size of the egg was noted. The oil drop had moved to an upper sector and the surrounding egg wall had become thin (Fig. 5). At 7:50 P.M. there was no further increase in size of the egg but the oil drop had become small, and the wall very thin. There was a general darkening of the cytoplasm due to increased granulation (Fig. 6). The oil drop completely disappeared by 8:15 P.M. and a central vacuole had become evident (Fig. 7).

A short germ tube grew from the egg during the approximate five hours between 8:15 P.M., June 5, and 1:05 A.M., June 6. The end of this tube was touching a pit on the oogonial wall. The granular cytoplasm became lighter and the egg had increased slightly in size (Fig. 8). By 3:10 A.M., June 6, the germ tube had grown through the pit to the outside. A definite constriction was noted as the tube passed through the pit and a swelling occurred immediately after opening to the outside. The cytoplasm was now quite pale and a vacuole could be seen in the egg and the germ tube (Fig. 9). At 5:10 A.M. this tube had grown to a distance of 138 micra and a cross wall had been formed cutting off a sporangium (Fig. 10). The old egg was by now completely empty. By 9:30 A.M. spores had emerged and were clustered at the mouth of the sporangium. These were later observed to germinate and swim away.

The germination process began four days and ten hours after the mycelium was placed in fresh water. Germination was completed twenty-four hours after it began.

Achlya proliferoides Coker

The culture of this form was eight weeks and six days old when placed in fresh water. It was kept under observation one week before any signs of germination were noted. Its development was exactly the same as that described for *A. flagellata*, which shows a further very close relationship between these two species. This experiment has since been repeated twice with the hope that some dissimilarity between the two species might be found which would provide a really distinctive key character. Development took thirty-one hours from the first encircling of the oil drop by the cytoplasm to the emptying of the seven spores from the germ tube sporangium.

Achlya caroliniana Coker

Plate 4, figure 11

This specimen of *A. caroliniana* was two months and five days old when it was placed in fresh water at 4:00 P.M., July 7, 1947. Its development closely resembled that of the other species of *Achlya* with eccentric eggs. In my culture germination was effected by the formation of a sporangium at the apical end of the germ tube (Fig. 11). Numerous eggs in the culture germinated and the average zoospore count of ten sporangia was seven.

The first evidence of germination was seen twenty hours after the beginning of the experiment and was completed within forty-eight hours after it began.

Achlya americana Humphrey

Plate 4, figures 12-13

The identification of this form is still in doubt as it somewhat resembles *A. conspicua* Coker. However, we will call it *A. americana* until a positive identification can be made. Several attempts had been made to germinate this species, which had been in a resting condition since May 5, 1947, but none was successful until a portion of the mycelium was placed in fresh water at 10:15 A.M. on September 29, 1947.

The development of the eccentric eggs closely parallels that described for *A. flagellata*. Germination is effected by the production of a germ tube with an apical sporangium (Fig. 12). However, it was not unusual for the growth of the tube to continue another twenty-four hours before a sporangium was produced. This growth resulted in a sparsely branched mycelium with an apical sporangium which, in general, was smaller than the one produced by a shorter tube (Fig. 13).

On the whole my observations made on this species follow those made by Trow (1899) on *A. americana* var. *cambrica*, although there seems to be a great divergence in the length of resting periods of these two forms.

Germination began twenty-four hours after the mycelium was placed in fresh water. It was complete three days and seven hours after it began.

Achlya glomerata Coker

Plate 5, figures 1-2

This form was three weeks and five days old when placed in fresh water at 4:15 P.M., October 3, 1947.

The first stage in germination—the surrounding of the oil globule by the cytoplasm—was seen at 11:15 A.M. on October 4. The changes in the zygote during the germination process were like those described for other species of *Achlya* with eccentric eggs. By 2:25 P.M., October 6, 1947, a germ tube about nine times the width of the zygote had formed and an apical sporangium had been cut off (Fig. 2). The sporangium passed through the usual changes prior to spore release and five spores were liberated at 3:45 P.M. the same afternoon.

However, only about one half of the zygotes in my culture germinated in the manner described above. Very frequently, the zygote produces a long branched mycelium which twines in and out among the other eggs in the culture. A single zygote was followed through in its development and it produced the mycelium shown in Figure 1. About ninety per cent of the zygotes on the slide germinated during this experiment.

The first evidence of germination was seen nineteen hours after the mycelium was placed in fresh water and required two days and four hours for completion.

Achlya crenulata Ziegler

Plate 5, figure 3

This form (Ziegler, 1948) was six weeks and three days old when placed in fresh water at 9:00 A.M., June 16, 1947.

The germination process is typical of those described for other species of *Achlya* with eccentric eggs. The egg that was followed through its development unfortunately did not show the typical condition in germ tube formation. As shown in Figure 3, it produced a germ tube which grew out some distance and then formed a branch near its base which also grew out. The cytoplasm of the original germ tube flowed into this secondary branch and formed a sporangium.

However, many eggs germinated in the culture and all, with this one exception, formed a single germ tube with an apical sporangium. The zoospores from ten different sporangia were counted and the average was nine.

Germination probably began at about the twenty-sixth hour after the mycelium was placed in fresh water and was completed within an additional two days and six hours.

Achlya recurva Cornu

Plate 5, figure 4

The oogonia of this species were two weeks and two days old when they were placed in fresh water.

The development of the germinating eggs of this species closely parallels those eccentric eggs such as *A. flagellata*, *A. proliferoides*, and *A. caroliniana* already described. The only difference we have been able to see is that the oil drop usually does not disintegrate entirely before germ tube formation but may persist—at least a small part of it—until the germ tube has attained some length.

Latham (1935) observed the germinating eggs of this species but did not follow the process through in detail. This species is very closely related to *Thraustotheca primoachlya* and resembles it not only in external features but also in the fact that its eggs germinate readily. It is distinguished from *T. primoachlya* by having the type of sporangia characteristic of *Achlya*, and longer protuberances. *T. primoachlya*, of course, produces sporangia of the *Thraustotheca* type. See Plate 5, figure 4, and Plate 6, figure 1, for comparison.

Achlya recurva began to germinate twelve hours after it was placed in fresh water and the process was completed in thirty-one hours after it began.

Achlya apiculata de Bary

Plate 5, figure 5

The culture of this form was nine weeks and five days old when it was placed in fresh water. Development resembles that described for *A. colorata*, *S. ferax*, and *I. unispora*. Germination is usually effected by the production of sporangia. However, the eggs do occasionally produce long mycelial threads which usually do not branch (Fig. 5).

Coker (1923) had germinated the eggs of *A. apiculata* var. *prolifera* on agar. On a solid medium, long unbranched threads were formed. However, as we have shown here, eggs germinated in water predominantly form sporangia, with an occasional thread formation.

Germination began five days and twenty-three hours after the mycelium was placed in fresh water. It was completed after an additional four days and four hours.

Protoachlya hypogyna Shanor and Con.*Achlya hypogyna* Coker and Pem.

Plate 5, figures 6-8

The nomenclature of this species is confused and misleading. In 1908 Coker and Pemberton (Bot. Gaz. 45: 194) described *Achlya hypogyna* from Chapel Hill. In 1923 Coker (The Saprolegniaceae, p. 90) established the genus *Protoachlya* and transferred to it his old species *A. paradoxa* but not *A. hypogyna*. In 1942 Shanor and Conover described a species from Illinois as *Protoachlya hypogyna* and attempted, on the basis of what was then known of *Achlya hypogyna*, to show that the two were different. At that time Dr. Coker agreed to the distinction. However, later culture work has proved beyond a doubt that the Chapel Hill species belongs to the genus *Protoachlya* and is the same as the plant from Illinois, but the combination *Protoachlya hypogyna* (Coker and Pem). Shanor and Con. cannot be used as the latter authors were not transferring the older species.

Miss Josephine Stewart collected *A. hypogyna* in Chapel Hill again in 1946 and it has been maintained in culture since that time. As Figures 6-8 show, the eggs on germinating produce long unbranched germ tubes with apical sporangia. A total of nine sporangia were seen and all discharged their spores in the manner typical of *Protoachlya*.

Germination began twenty hours after the mycelium was placed in fresh water. It was completed within seventy-two hours after it began.

Aphanomyces laevis de Bary

Plate 5, figure 11

Several previous attempts to germinate the eggs of this species resulted in failure but germination was seen after the culture was five months and two weeks old. The development of the eggs recall that of *A. flagellata* and other species of *Achlya* with eccentric eggs. Three types of germination were found. In the first, the germ tube formed a long, unbranched hypha. In another type, which

was by far the most common seen in my preparations, the original germ tube branched after leaving the oogonium and finally produced a small, branched mycelium. A low power view of this condition may be seen in Figure 11. The last type was seen only twice. It consisted essentially of the mycelium plus a single sporangium.

As recorded under "Historical Notes", several other workers have seen the germinating eggs of this genus, among them, de Bary, Sorokine, Kasanowsky, and Jones and Drechsler.

Twenty-three hours after being placed in fresh water the first sign of germination was seen. The process was completed in about three days and twenty-one hours.

Thraustotheca clavata (de Bary) Humphrey

Plate 5, figures 9-10

The culture of this form was eleven weeks and three days old when a portion of the mycelium was placed in fresh water.

My observations on the germination of this specimen closely follow Weston's (1918) descriptions and drawings in his work on this species. However, as only pure water was used in my cultures, no extensive mycelial formation occurred. Weston's Figure 59 shows what he describes as an oil globule becoming irregular and disappearing. This closely resembles the disintegrating eggs which are quite common throughout a culture. The gradual absorption of the oil drop is very easily seen in fresh water mounts and does not differ from the process described in *A. flagellata*. Sporangial formation is shown in Figures 9 and 10.

Germination began four days and four hours after the mycelium was placed in fresh water. The process was completed five days after it began.

Thraustotheca primoachlya Coker and Couch

Plate 6, figure 1

This species and *Dictyuchus missouriensis* were sent to me by Dr. Leland Shanor of the University of Illinois. The culture was four and one half weeks old when placed in fresh water. The stages of egg germination in this species closely resemble those already described for *T. clavata*, and *A. flagellata*. Coker and Couch (1924) have already worked out the details of egg germination for this species. Germination in this form readily occurs after a rest period of at least ten days. I was not able to find any eggs in which the entire tube and egg space became the sporangium.

Twenty-five hours after the mycelium was placed in fresh water the first stage in germination was noticed. An additional twenty-six hours was needed for completing the process.

Dictyuchus missouriensis Couch

Plate 6, figures 2-7

The culture of this form was fourteen weeks and one day old when it was placed in fresh water at 9:45 A.M., October 6, 1947 (Fig. 2). The first visible

evidence of germination was noticed at 12:15 P.M., October 8, when the diminishing oil globule was surrounded by granular cytoplasm (Fig. 3).

The germination stages for this species are like those described and drawn by Couch (1926) in his work on heterothallism in this genus. The egg walls become irregular and scalloped, the oil droplet is broken up, and a germ tube is produced. Figures 3 to 5 show these stages.

The germ tubes of this species are usually from ten to thirteen times longer than the oogonial width (Fig. 7). Those seen by Couch appear to be much shorter. From six to eleven spores are produced in the sporangia.

The first evidence of germination was noted two days and three hours after the mycelium was placed in fresh water. The germination process required two days and twenty-three hours.

Geolegnia inflata Coker and Harvey

Plate 6, figures 17-23

This species and the following one were collected by Mr. William Pendergrass in soil collections from Florence, S. C.

The mycelium was six weeks and two days old when placed in fresh water at 9:30 A.M., September 29, 1947 (Fig. 17). Due to the extreme smallness of the species, the first stages in germination were missed but by 1:45 P.M., October 1, the oil globule had come to occupy a more central position and had decreased to about one half its original size (Fig. 18). By 4:35 P.M. a further decrease in size of the oil drop was noted and the wall appeared scalloped on the inner side. There had been a gradual darkening of the cytoplasm during the interval (Fig. 19).

At 9:00 A.M., October 2, the germinating zygote had increased in size and the wall had become thin. A central vacuole had developed (Fig. 20). A germ tube began to protrude from the zygote at 11:10 A.M., October 2, and by 12:10 P.M.; October 3, this germ tube had attained the length shown in Figure 21.

Geolegnia inflata will germinate readily after a rest period when it is placed in fresh water. The germ tubes form a primary mycelium without branches and I have never seen spores formed in this species. The lengths of two of these germ tubes are shown in Figures 22 and 23.

The germination of this species closely resembles those described for the eccentric *Achlya* species although its size somewhat restricts careful observation.

The germination process began forty-eight hours after the mycelium was placed in fresh water. It was completed within two days and three hours after it began.

Brevilegnia linearis Coker and Braxton

Plate 6, figures 8-16

My culture of this form was three weeks old when it was placed in fresh water at 10:50 A.M., July 17, 1947 (Fig. 8).

Details of the germination process were extremely difficult to see. The very small oogonia, each with a single egg, are the same size as the abundant zoospores

and the eggs are frequently to be seen without the oogonial wall and intermingled with the zoospores.

The first stage in germination was not noticed until 12:10 P.M., July 19, when the cytoplasm became slightly darker and surrounded the fat globule (Fig. 9). By 8:25 P.M. the same day, the wall had begun to thin and the oil droplet had decreased noticeably in size (Fig. 10). On July 20 at 11:25 A.M., the cytoplasm had become very granular, the oil drop had further decreased in size, and a slight enlargement of the egg was noticed (Fig. 11). By 7:50 P.M. the same day, a vacuole had developed in the egg and the oil globule was much smaller (Fig. 12).

At 8:15 A.M., July 21, a small germ tube had arisen and protruded through the oogonial wall, probably through a thin place in the wall caused by the antheridial tube (Fig. 13). By 9:25 A.M., July 22, this germ tube had attained a length of about ten times the diameter of the oogonium but did not branch (Figs. 14-15).

In eight germinating eggs observed in my cultures, three germinated in the manner described above with a long unbranched hypha. Five of the eight eggs had long branching germ tubes such as is shown in Figure 16. No sporangia were observed.

Germination began twenty-five hours after the mycelium was placed in fresh water and required an additional two days and twenty hours for completion.

TESTS FOR FATTY MATERIALS

In the eggs of the Saprolegniaceae, numerous small droplets or one large droplet of a substance which has been called oil or fatty reserve are present. These droplets give the eggs their characteristic appearance; *e.g.*, a centric egg has one or two layers of small oil droplets completely surrounding the cytoplasm; a sub-centric egg has two or more layers on one side and one layer on the other; an eccentric egg has a single large drop completely outside the protoplasm. In order to determine the exact composition of this material, several tests have been performed. These tests have certainly been made by other workers in this field but a search of the available literature has failed to disclose any mention of them in the articles which I have examined. They are:

1. Sudan III. Used as a presumptive test. Stains all fatty substances deep orange yellow.
2. Sudan IV (Scarlach R). Stains fats orange red.
3. Osmic acid. Exposure to osmic acid 1/2 % solution for several hours usually completely blackens all fats.
4. Nile blue sulphate. Although this is a controversial stain in that it is doubtful whether a stain can differentiate between such components as fatty acids, mixtures of fats, and true fats, we have included it here simply for a record. With this stain, fatty acids become pink, mixtures of fats turn violet, and fats become blue.
5. Saponification. 20% NH_4OH and saturated aqueous KOH. When material is treated with equal parts of the above mixture and then observed under polarized light, sphaerocrystals should become evident if fat is present.

6. Polarization. Fats under polarized light should have an isotropic appearance if no fat-like substances are present.

7. Solubilities. Fats are soluble in ether, petrol ether, chloroform, acetone, benzene, carbon disulphide, carbon tetrachloride, phenol, xylol and other substances. Several of these solvents were applied to the fungus material.

The results were as follows:

1. Sudan III

A. flagellata—large oil drop stained dark orange.

A. apiculata—oil droplets stained bright orange.

S. ferax—oil droplets stained bright orange.

2. Sudan IV

A. flagellata—large oil drop stained orange yellow.

A. apiculata—oil droplets stained orange yellow.

S. ferax—oil droplets stained orange yellow.

3. Osmic acid. Exposed to fumes of 1/2% for two hours. Oil in all cases turned black.

4. Nile blue sulphate. Oil in all three cases stained blue with no indication of other colors.

5. Saponification. Under treatment described above, numerous sphaerocrystals were observed in all three species.

6. Polarization. All oil droplets were isotropic.

7. Solubilities. Solubility tests were made using xylol, ether, acetone, and carbon tetrachloride. In all cases, the fatty material was either soluble or partly soluble.

The results indicate that the material present in the three forms studied is true fat and that no fat-like substances are present. The slight divergence of the results in the solubility tests is due, perhaps, to the presence of some additional substances which are not fatty.

EFFECTS OF pH ON GERMINATION

Various factors such as light, temperature, amount of food, and pH of the medium influence the germination process in the eggs of the Saprolegniaceae. An experiment to determine the role of pH on the eggs of several species is described here. Five species representing several genera and both eccentric and centric eggs were used. These were *Protoachlya hypogyna* (centric eggs), *Saprolegnia ferax* (centric eggs), *Saprolegnia delica* (centric eggs), *Achlya recurva* (eccentric eggs), and *Thraustotheca primoachlya* (eccentric eggs).

The various pH solutions were made by using different amounts of a 0.1 molar solution of KH_2PO_4 and K_2HPO_4 and were checked on an electric potentiometer. The cultures were mounted in the usual manner but the pH solution was used instead of fresh water. After seventy-two hours, the results were studied. This experiment has been repeated several times with comparable results being received each time.

Several conclusions were reached after a study of the data obtained. In all cases a low pH such as 4.9 and 5.9 tended to inhibit the germination of the zygotes, as very few would germinate. If they germinated at this pH range,

they deviated from their usual germination pattern in fresh water. In most cases where they germinated by germ tubes with apical sporangia, a low pH caused the eggs to produce long, branched germ tubes with no formation of sporangia. Except in the case of *Achyla recurva*, zygotes would not germinate in a pH of 7.9. No zygotes germinated in a solution of 8.8. The zygotes underwent the same development in a pH of 6.9 as they do in fresh water.

EFFECT OF LIGHT ON GERMINATION

Sorokine, in 1876, following the work of Anton de Bary, was able to germinate the eggs of *Aphanomyces*. He discovered, among other facts, that the eggs would germinate only if they remained in the light. In reviewing the literature, this seems to be the only reference to this phenomenon. In order to corroborate this fact, three species which I have found to germinate readily were chosen. They were *Protoachlya hypogyna*, *Achyla recurva* and *Thraustotheca primoachlya*.

These forms were placed on slides in the manner described under "Materials and Methods." Two cultures were made of each species. One culture of each was immediately placed in the dark in the desk and the other placed on the laboratory table. Thermometers showed that the temperature on the table and in the desk were the same. (The temperature was not kept constant but the variations were the same.) Water was changed on each culture every morning. The cultures were opened and examined at the end of seventy-two hours and results recorded. The culture kept in the dark was then left in the light for an additional seventy-two-hour period to see if the zygotes would germinate.

The results can easily be summarized. For the three species used in this experiment, light is necessary for germination. This experiment has been repeated seven times and the results each time were comparable to each other. Occasionally, several zygotes have germinated in the dark but their total average would scarcely be one-sixth per ten fields.

After the above had been demonstrated, several trials to test egg germinations under different wave lengths of light were made. However, the results were inconclusive as too many variable factors were present.

A COMPARISON OF THE TYPES OF RESTING BODY GERMINATION IN THE OOMYCETES

This thesis is chiefly concerned with the germination processes exhibited by the members of one family, the Saprolegniaceae. The germination of the resting bodies of other families and orders of the Phycomycetes has been seen from time to time but the references to these phenomena are scattered throughout the various texts and sundry papers.

We have therefore endeavored to bring together the forms in which germination has been seen and attempt to fit them into different groups or patterns of germination. Finally, we shall try to make several general remarks concerning germination in this entire class.

As will be seen in the following pages, the germination patterns of the resting

bodies of the Phycomycetes are highly varied and these will be discussed by taking up each order separately.

Chytridiales

Sparrow (1943) lists and describes many species of Chytrids. The resting bodies of a great many of these forms have never been seen. In only 24 species of Chytrids has the germinating process been witnessed.

The resting spore is formed sexually in but 5 of the 26 species which have been seen to germinate. These are: *Polyphagus euglenae*, *Rhizophydium Couchii*, *R. ovatum*, *Micromyces longispinosus*, *Pringsheimiella dioica*, *Synchytrium endobioticum*, and possibly *Diplophlyctis intestina*.

Due to the incompleteness of the literature, it would be impossible to make any general conclusions regarding resting spore germination in the whole order. However, the forms in which germination has been seen do fall into several distinct groups. These are:

1. Those in which a small apical pore is formed with the resting body functioning as a pro-sporangium. The sporangium formed may or may not have an operculum.

Rhizidium mycophilum, *Polyphagus euglenae*, *Rhizophydium Couchii*, *Entophlyctis vaucheriae*, *Endochytrium* sp., *Diplophlyctis intestina*, *Megachytrium westonii*, *Chytrium olla*, *C. schenkii*, *C. lagenaria*, *Rhizidiopsis emmanuelensis*, *Phlyctochytrium synchytrii*, *Micromyces longispinosus*, *Blyttomyces spinulosus*, *Rhizosiphon crassum*, *Septochytrium variabile*.

2. Those in which the resting body functions as a sporangium forming a discharge tube.

Diplophlyctis intestina, *Rhizophydium ovatum*, *R. transversum*, *R. messanense*, *Olpidium luxurians*, *Pringsheimiella dioica*, *Rozella allomycis*, *R. achlyae*, *Cladochytrium replicatum*.

3. This group includes only one form: *Zygochytrium aurantiacum*. In this group the exospore wall cracks and the endospore wall elongates into a thick tube.

Sparrow (1943) points out "... Sorokin's genus has been almost universally rejected by monographers and mycologists in general. This rejection has been strengthened by the lack of further observations on the organism. Sorokin's descriptions and figures of the development and reproductive processes are unusually clear and distinct, and there seems no reason for doubting them. Since it is now apparent that the chytrids have evolved a variety of types of sexual reproduction it is not surprising to find a form having a method like that observed in another group of Phycomycetes." Sparrow is referring, of course, to the similarities between the sexual reproduction of this form and that, say, of *Rhizopus*. However, the germination of this form indicates a relationship with the oomycetes.

The changes in the resting body prior to germination are described by Karling (1939). These are:

1. Small germ tube pore develops in the thick wall.

2. Meanwhile the refractive central globule changes shape.
3. Protoplasm begins to ooze through the pore.
4. Globule becomes invaded on the periphery adjacent to the pore.
5. Sporangium develops in the outside; main globule now broken up.
6. Spores cut out.

It will be noted that several of the steps are reminiscent of those found in the Saprolegniaceae. For example, the breaking up of the oil globule, the passage of cytoplasm to the outside before the oil is completely disintegrated as in *A. recurva*, and the cutting out of the spores in the sporangium on the exterior of the original resting spore.

Blastocladales

The resting body of the Blastocladales is a thick-walled, punctate structure which is a sporangium in function and which liberates zoospores on discharge. It is comparable to the resting body of the Chytrids but not to the resting bodies of the higher forms.

As stated before, in this Order, germination of the resting spore always produces zoospores. Some other criterion must be found, therefore, in order to attempt any classification of the species in which germination has been seen. This criterion might possibly be the manner in which the resting spore wall cracks open. It is unfortunate, though, that this event has not been sufficiently described in some of the literature. If this characteristic can be used, the species fall into the following types:

1. Those in which the resting spore germinates by the cracking of the exospore wall into several irregular pieces after which the hyaline inner part sends out one or more papillae. (A key character in the diagnosis of *Blastocladia*--see Couch and Whiffen, 1942.)

Blastocladia variabilis, *B. stübenii*, *B. simplex*, *B. asperosperma*, *B. lacvisperma*, *B. microcystogena*.

2. Those in which the resting spore wall splits open along a pre-formed longitudinal line.

Blastocladia pringsheimii (not actually stated but drawings show this to be highly probable), *Coelomomyces* sp.

3. That one in which the outer wall of the resting body cracks and a discharge tube is formed, the resting body then functioning as a sporangium.

Catenaria anguillulae.

4. Those in which the resting body germinates to form cystospores, each of which gives rise to four uninucleate gametes which emerge through a pore.

Catenaria allomyces, *Blastocladia cystogena*, *Allomyces cystogenus*.

5. Those in which the wall cracks open irregularly, papillae are formed, and zoospores are liberated; and those in which no details of wall cracking are available.

Allomyces arbusculus, *A. javanicus*, *Blastocladia parva*.

Monoblepharidales

Germination in this Order has been seen in five species belonging to two genera.

Where it has been noted, the resting period of the zygote before germination is about a month. These five species appear to fall into two groups:

1. Those in which the resting spore germinates by the cracking of the oospore wall and the production of a hypha or a mycelium (Sparrow, 1933).

Monoblepharis macrandra, *M. polymorpha*, *Monoblepharella taylori*, *M. mexicana*.

2. That one in which a hypha passes to the outside through a pore in the oogonial wall and then forms a mycelium.

Monoblepharella elongata.

It is impossible to compare this group with the Blastocladales as no details of the wall cracking seem to have been recorded. Also there appears to be no recorded stage in the germination process.

Of the species mentioned above, *M. polymorpha* germinates by producing a hypha. It may possibly be that this would later develop into a mycelium.

Hypochytriales

Only one example of germination was found in this Order. In *Hypochytrium hydrodictii* the resting body forms a single discharge papilla through which the spores are liberated.

Leptomitales

There are three examples of germinating oospores in this Order. In all three examples, the germinating oospore produces one or more slender hyphae. These forms are:

Apodachlya brachynema, *A. minima*, *Apodachlyella completa*.

The stages of germination have been observed by Coker and Leitner (1938) in *A. minima*:

1. Cytoplasm becomes irregular in outline where pressed against the oil droplet and a few patches of small granules appear in the hyaline cytoplasm.
2. Cytoplasm becomes granular throughout.
3. Egg enlarges and splits oogonial wall.
4. One or two germ tubes formed which fill with cytoplasm.
5. Meanwhile, oil globule diminishes in size and is apparently used up.
6. When egg is empty, backflow into egg is prevented by the formation of a plug.

Several of these stages are found in the germinating resting bodies of other Orders. The breaking down of the oil globules and the darkening cytoplasm recall the series of events in the Chytridiales and the Saprolegniales.

Lagenidiales

There are four examples of germinating bodies in this Order. These are: *Olpidiopsis achlyae*, *O. varians*, *O. schenkiana*, *Lagenidium rabenhorstii*.

In the first three of the examples, the resting body functions as a sporangium with a short germination tube from which several spores are discharged. The last case, *L. rabenhorstii*, is rather unique. Twenty-four hours after wall formation on the resting spore, it breaks down and liberates a single spherical zoospore. However, as Sparrow (1943) says, "Although the quick germination of resting

structures is recorded in other fungi, the 'breaking down' of the wall and, in particular, the highly inefficient process of liberating only a single zoospore from a sexually formed structure are rare, if not unknown, among these plants and need confirmation."

No details of the germination process for any of the species have been found.

Peronosporales

Several examples of resting spore germinations are recorded for this Order. The species seem to fall into the following four groups:

1. That one in which the egg, after resorption of its wall and oil globule, functions as a sporangium.

Pythiogeton utriforme.

2. Those in which the oospore forms one or more branched hyphae.

Pythiomorpha gonapodyides, *Pythium adhaerens*, *P. angustatum*, *P. ultimum*.

3. Those in which the oospore forms a germ tube with an apical sporangium, or, certain branches of the germ tube form sporangia.

Pythium adhaerens, *P. angustatum*, *P. dictyosporum*.

4. That one in which the epispore and surrounding colorless membrane breaks irregularly and the endospore bulges out, forming a sporangium.

Cystopus candidus.

Whenever the stages of germination were noted they agree in the following facts (Trow, 1901; Sparrow, 1931):

1. The inner wall of the oospore was dissolved.

2. The fatty mass was broken up.

3. The protoplasm increased in amount.

4. One or more germ tubes arose from the germinating eggs.

The same stages named above are also found in the germinating resting bodies of the Chytridiales and Saprolegniales.

GENERAL REMARKS ON GERMINATION IN THE PHYCOMYCETES

From what has been presented on the preceding pages, certain general statements can be made concerning germination in this class of fungi.

1. The outer wall of the resting body is pierced by a long or short discharge tube, or cracked during the germination process.

2. The inner layer of the resting body or the thinned zygote wall protrudes through the outside wall either in the form of papillae or germination tubes.

3. Whenever fatty reserve is found in the resting body, either in one single drop, or several globules, it is always broken up into recognizable smaller particles or disintegrated.

4. In general, the following types of germination are seen in the Phycomycetes:

A. Those in which the resting body is transformed into a sporangium which discharges spores.

Chytridiales:—*Diplophlyctis intestina*, *Rhizophyidium ovatum*, *R. transversum*, *R. messanense*, *Olpidium luxurians*, *Pringsheimiella dioica*, *Rozella alomycis*, *R. achlyae*, *Cladochytrium replicatum*.

Blastocladales:—*Blastocladiella* sp., *Blastocladia pringsheimii*, *B. parva*, *Coelomomyces* sp., *Catenaria anguillulae*, *C. allomyces*, *Allomyces* sp.

Hypochytriales:—*Hypochytrium hydrodictii*.

Saprolegniales:—*Achlya flagellata*, *A. orion*.

Lagenidiales:—*Olpidiopsis achlyae*, *O. varians*, *O. schenkiana*.

Peronosporales:—*Pythiogeton utrifforme*.

B. Those in which the resting body functions as a prosporangium.

Chytridiales:—*Rhizidium mycophilum*, *Polyphagus euglenae*, *Rhizophydium Couchii*, *Entophlyctis vaucheriae*, *Endochytrium* sp., *Diplophlyctis intestina*, *Megachytrium westonii*, *Chytrium olla*, *C. schenkii*, *C. lagenaria*, *Rhizidiopsis emmanuelensis*, *Phlyctochytrium synchytrii*, *Micromyces longispinosus*, *Blytiomyces spinulosus*, *Rhizosiphon crassum*, *Septochytrium variabile*.

Peronosporales:—*Cystopus candidus*.

C. Those in which the resting body forms a short or long germ tube with apical sporangium (ia), or, a branched tube with certain branches forming sporangia.

Saprolegniales:—*Aplanes treleaseanus*, *Saprolegnia ferax*, *S. delica*, *S. megasperma*, *S. dichina*, *S. litoralis*, *Achlya colorata*, *A. racemosa*, *A. flagellata*, *A. proliferoides*, *A. americana*, *A. caroliniana*, *A. apiculata*, *A. recurva*, *A. glomerata*, *A. crenulata*, *Isoachlya itoana*, *I. unispora*, *Aphanomyces laevis*.

Peronosporales:—*Pythium adhaerens*, *P. angustatum*, *P. dictyosporum*.

D. Those in which the resting body forms a long or short germ tube, or a branching mycelium. •

Chytridiales:—*Zygocytrium aurantiacum*.

Monoblepharidales:—*Monoblepharis macrandra*, *M. polymorpha*, *Monoblepharella taylori*, *M. mexicana*, *M. elongata*.

Saprolegniales:—*Isoachlya unispora*, *I. intermedia*, *Aphanomyces laevis*, *Brevilegnia linearis*, *Geolegnia inflata*, *Achlya glomerata*.

Peronosporales:—*Pythium adhaerens*, *P. angustatum*, *P. ultimum*, *Pythiomorpha gonapodyides*.

The writer made the above study in an endeavor to ascertain whether any phylogenetic relationships between the members of the Saprolegniaceae and other Phycomycetes could be made out. As can be seen the results are inconclusive. It can be pointed out, however, that as a rule the wall of the resting spore in the Chytrids and Saprolegniales is digested internally and a pore or a germ tube is formed, while the frangible wall of the Blastocladales is generally cracked.

SUMMARY

The zygotes of twenty-six species of the Saprolegniaceae have been germinated and their germination processes followed through in detail. Of these twenty-six, only four have been followed through in some detail by other observers. Six other observers have mentioned seeing the germinated zygote in six species but no details of development were given. The zygotes of the Saprolegniaceae will germinate after a rest period if the mycelium is placed in fresh

charcoal distilled water. It was necessary to employ a special decoction (corn-grain juice) in only two species of the twenty-six, *I. itoana* and *I. intermedia*.

The germinated zygotes fall into four types:

A. Those in which a long or short germ tube is formed with an apical sporangium. The following forms are included in this type: *Aplanes treleaseanus*, *Saprolegnia delica*, *S. ferax*, *S. megasperma*, *S. dictina*, *S. litoralis*, *Achlya colorata*, *A. racemosa*, *A. flagellata*, *A. proliferoides*, *A. americana*, *A. caroliniana*, *A. apiculata*, *A. recurva*, *A. glomerata*, *A. crenulata*.

B. Those in which the germ tube produces a sparsely branched mycelium with a sporangium at the apex of the main hypha or a branch. This type includes: *Isoachlya unispora*, *I. intermedia*, *I. itoana*, *Achlya americana*, *Aphanomyces laevis*.

C. Those in which the primary germ tube forms a branched mycelium. This group includes: *Isoachlya unispora*, *I. intermedia*, *Achlya glomerata*, *Aphanomyces laevis*, *Brevilegnia linearis*.

D. Those in which the primary germ tube forms a long unbranched hypha. These are: *Aphanomyces laevis*, *Brevilegnia linearis*, *Geolegnia inflata*.

The food material contained within the egg has been proved to be true fat in *Achlya flagellata*, *A. apiculata*, and *Saprolegnia ferax*.

Experiments involving the effects of pH on germination have demonstrated that low pH's and alkaline solutions tend to inhibit germination; that the low pH ranges tend to cause the germinating eggs to deviate from their usual development in fresh water; and that eggs in a pH of 6.9 undergo the same development as they do in fresh water.

An experiment, repeated several times, on the effect of light on germination has demonstrated that at least for the three species used in the experiment light is necessary for germination.

A study of the germinating resting bodies of other Phycomycetes has been made and these show various patterns of germination. These patterns, however, show no phylogenetic connections between the members of the Saprolegniaceae and other Phycomycetes as far as resting body germination is concerned.

It has been shown that the zygotes of the Saprolegniaceae can be germinated easily. These forms are large and easily grown. It is suggested, therefore, that they might lend themselves readily to experiments involving x-ray, exposure to ultra-violet light, and general genetical studies.

ACKNOWLEDGMENTS

I am deeply indebted to Dr. J. N. Couch, who suggested this problem, for his aid and encouragement during this study and to Dr. W. C. Coker for his continued interest. I also wish to thank Mrs. C. D. Beers for help in arranging the plates.

LITERATURE CITED

- COKER, W. C. 1923 The Saprolegniaceae. University of N. C. Press. Chapel Hill, N. C.
— AND COUCH, J. N. 1924 Revision of the Genus *Thraustotheca* with a Description of a New Species. Jour. Elisha Mitch. Sci. Soc. 40: 197-202.

- AND LEITNER, JANE. 1938 New Species of *Achlya* and *Apodachlya*. Jour. Elisha Mitch. Sci. Soc. **54**: 311-318.
- AND MATTHEWS, V. D., AND [bibliog.] BARNHART, J. H. 1937 North American Flora II: part 1.
- COUCH, J. N. 1926 Heterothallism in *Dictyuchus*, a genus of the water molds. Ann. Bot. **40**: 849-881.
- AND WHIFFEN, A. J. 1942 Observations on the Genus *Blastocladiella*. Amer. Jour. Bot. **29**: 582-591.
- DE BARY, ANTON. 1860 Einige neue Saprolegnieen. Jahr. f. Bot. **2**: 169-192.
- 1863 Recherches sur le Developpement de Quelques Champignons. Ann. d. Sci. Nat., 4 Ser. **20**: 1-148.
- 1884 Comparative Morphology and Biology of the Fungi, Mycetozoa, and Bacteria. English Edition. Clarendon Press. Oxford.
- 1888 Species der Saprolegnieen. Bot. Zeitung **46**: 646-659.
- FITZPATRICK, H. M. 1930 The Lower Fungi. McGraw-Hill Book Co. Inc., New York.
- INDOH, H. 1939 Studies of the Japanese Aquatic Fungi. I. On *Apodachlyella completa* sp. nov. with a Revision of the Leptomitaceae. Science Reports Tokyo Bunrika Daigaku **4**: no. 66, 43-50.
- JONES, R. J. AND DRECHSLER, C. 1925 Root Rot of Peas in the U. S. Caused by *Aphanomyces euteiches* (N. Sp.). Jour. Agric. Res. **30**: no. 4, 293-325.
- KARLING, J. S. 1939 Studies of *Rhizophidium*. III. Germination of the Resting Spores. Bull. Torrey Bot. Club **66**: 281-286.
- KASANOWSKY, V. 1911 *Aphanomyces laevis* de Bary. I. Entwicklung der Sexualorgane und Befruchtung. Ber. d. Deutsch. Bot. Ges. **29**: 210-228.
- KLEBS, GEORGE. 1899 Zur Physiologie der Fortpflanzung einiger Pilze. II. *Saprolegnia mixta* de Bary. Jahr. f. Bot. **33**: 513-593.
- LATHAM, D. H. 1935 *Achlya recurva* Cornu from North Carolina. Jour. Elisha Mitch. Sci. Soc. **51**: 152-156.
- LEE, B. 1946 The Microtometist's Vade-Mecum. 10th Ed. The Blakiston Co., Philadelphia, Pa.
- LAIBACH, F. 1926 Zur Cytologie von *Monoblepharis*. Ber. d. Deutsch. Bot. Ges. **44**: 59-63.
- MATTHEWS, V. D. 1931 Studies on the Genus *Pythium*. University of N. C. Press, Chapel Hill, N. C.
- PATTERSON, P. M. 1927 Fertilization in *Achlya colorata*. Jour. Elisha Mitch. Sci. Soc. **42**: 108-120.
- PRINGSHEIM, N. 1873-1874 Weitere Nachträge zur Morphologie und Systematik der Saprolegnieen. Jahr. f. Bot. **9**: 191-234.
- RAWLINS, T. E. 1933 Phytopathological and Botanical Research Methods. John Wiley and Sons, New York, N. Y.
- SCHLOSSER, L. A. 1929 Geschlechtsverteilung und Fakultative Parthenogenese bei Saprolegniaceen. Planta Archiv. f. Wiss. Bot. **8**: 529-570.
- SCHRADER, E. 1937-1938 Die Entwicklung von *Thraustotheca clavata*. Flora N. F. **32**: 125-150.
- SHANOR, L. 1942 A New *Monoblepharella* from Mexico. Mycologia **34**: 241-247.
- and Conover, Robert A. 1942 A New *Protoachlya*. Amer. Mid. Nat. **28**: 746-751.
- SMITH, G. M. 1938 Cryptogamic Botany Vol. 1. McGraw-Hill Book Co., Inc., New York, N. Y.
- SOROKINE, N. 1876 Quelques Mots sur le Developpement de *L'Aphanomyces stellatus*. Ann. d. Sci. Nat., 6 Ser. **3**: 46-52.
- SPARROW, F. K. 1931 Two New Species of *Pythium* Parasitic in Green Algae. Ann. Bot. **45**: 257-277.
- 1931 Observations on *Pythium dictyosporum*. Mycologia **23**: 191-203.
- 1933 The Monoblepharidales. Ann. Bot. **47**: 517-542.

- 1943 Aquatic Phycomycetes. University of Michigan Press. Ann Arbor, Michigan.
- SPRINGER, M. E. 1945 Two New Species of *Monoblepharella*. Mycologia 37: 205-216.
- TROW, A. H. 1895 The Karyology of *Saprolegnia*. Ann. Bot. 4: 609-650.
- 1899 Observations on the Biology and Cytology of a new variety of *Achlya americana*. Ann. Bot. 13: 131-179.
- 1901 Observations on the Biology and Cytology of *Pythium ultimum* n. sp. Ann. Bot. 15: 269-312.
- WAGER, H. 1896 On the Structure and Reproduction of *Cystopus candidus* Lév. Ann. Bot. 10: 295-342.
- WESTON, W. H. 1918 The Development of *Thraustotheca*, a Peculiar Water-Mould. Ann. Bot. 32: 156-170.
- WHIFFEN, A. J. 1943 New Species of *Nowakowskiella* and *Blastocladia*. Jour. Elisha Mitch. Sci. Soc. 59: 37-43.
- 1946 Two New Terricolous Phycomycetes belonging to the Genera *Lagenidium* and *Blastocladia*. Jour. Elisha Mitch. Sci. Soc. 62: 54-58.
- WOLF, F. A. AND WOLF, F. T. 1947 The Fungi. Vols. 1, 2. John Wiley and Sons, Inc. New York, N. Y.
- WOLK, F. T. 1944 The Aquatic Oomycetes of Wisconsin. Part 1. University of Wisconsin Press. Madison, Wisconsin.
- ZIEGLER, A. W. 1948 A New Species of *Achlya*. Mycologia. May-June Issue 1948—forthcoming.

EXPLANATION OF PLATES

Figures made with the aid of a camera lucida; arm at 120 mm., and mirror at 45°, with paper at table level. Magnification 1128 × unless otherwise indicated. Drawings reduced about one half.

PLATE 1

Aplanes treleaseanus (Humphrey) Coker.

Fig. 1. Oogonium with five eggs.

Figs. 2-6. Stages in germination.

Fig. 7. Germ tube with apical sporangium.

Fig. 8. Same, but with spores delimited.

Fig. 9. Low power view, 178 ×, of another oogonium showing the germinated eggs.

Saprolegnia ferax (Gruith.) Thuret.

Fig. 10. Oogonium, three eggs disintegrating.

Figs. 11-14. Stages in germination.

Fig. 15. Germ tubes from two germinating eggs passing to outside through oogonial wall pits.

Fig. 16. Empty sporangia of the above.

PLATE 2

Fig. 1. *Saprolegnia megasperma* Coker. Germinated egg with germ tube and apical sporangium.

Fig. 2. Same. Three sporangia formed. Note absence of cross wall.

Fig. 3. *Saprolegnia litoralis* Coker. Germinated eggs with germ tubes and apical sporangia.

Fig. 4. *Saprolegnia delicata* Coker. Oogonium in which four eggs have germinated. Note germ tubes and sporangia.

Fig. 5. *Saprolegnia diclina* Humphrey. Germinated eggs with germ tubes.

Fig. 6. Same. Empty sporangium.

Isoachlya unispora Coker and Couch.

Fig. 7. Oogonium with egg.

Figs. 8-10. Stages in germination.

Fig. 11. Germ tube has passed through oogonial wall pit to outside.

PLATE 3

Fig. 1. *Isoachlya unispora* Coker and Couch. Germinated egg with long germ tube and apical sporangium. Note proliferation of sporangia.

Fig. 2. *Isoachlya intermedia* (Coker and Harv.) Coker. Germinated egg with long germ tube showing sporangium formation and sparsely branched mycelium; oogonium in outline.

Achlya colorata Patterson.

Fig. 3. Oogonium with three eggs.

Figs. 4-6. Stages in germination.

Fig. 7. Beginning of germ tube.

Fig. 8. Germ tube passing to outside.

PLATE 4

Fig. 1. *Achlya colorata* Patterson. Germinated egg showing germ tube with apical sporangium.

Fig. 2. *Achlya racemosa* Hildebrand. Germinated egg with germ tube and apical sporangium which has discharged spores.

Achlya flagellata Coker.

Fig. 3. Oogonium with three eggs.

Figs. 4-7. Stages in germination.

Fig. 8. Beginning of germ tube.

Fig. 9. Germ tube passing through oogonial wall. Note constriction of the tube as it passes through the wall.

Fig. 10. Germinated egg showing long germ tube and apical sporangium.

Fig. 11. *Achlya caroliniana* Coker. Germinated eggs showing long germ tubes with sporangia.

Achlya americana Humphrey.

Fig. 12. Germ tube from a germinated egg of *A. americana* which has produced an apical sporangium.

Fig. 13. Same. Low power view, 85 X, of a germinated egg which has formed a sparsely branched mycelium with an apical sporangium at the end of the main hypha.

PLATE 5

Fig. 1. *Achlya glomerata* Coker. Low power view, 178 X, of an egg which has germinated to produce a long branched mycelium.

Fig. 2. Same. Another egg which has germinated to produce a long germ tube with an apical sporangium.

Fig. 3. *Achlya crenulata* Ziegler. Germinated eggs which have produced germ tubes. The one on the right has formed an apical sporangium.

Fig. 4. *Achlya recurva* Cornu. Germinated egg which has produced a long germ tube. End of germ tube is swelling preparatory to sporangium formation.

Fig. 5. *Achlya apiculata* de Bary. Low power view, 178 X, of an oogonium in which four eggs have germinated. Three have formed germ tubes with sporangia, while the fourth has produced a long unbranched hypha.

Protoachlya hypogyna Shanor and Con.

Fig. 6. Two germinated eggs from a single oogonium showing the germ tubes leaving the oogonium.

Figs. 7-8. The ends of the germ tubes shown in figure 6. The tubes were extremely long and would extend the full length of the plate before the portions shown here would be formed.

- Fig. 9. *Thraustotheca clavata* (de Bary) Humphrey. A germinated egg showing the germ tube with cytoplasm collecting in the far end.
Fig. 10. Same. The completed sporangium.
Fig. 11. *Aphanomyces laevis* de Bary. A single egg has germinated forming a long branched mycelium. 85 \times .

PLATE 6

- Fig. 1. *Thraustotheca primoachlya* Coker and Couch. Germinated egg with germ tube and apical sporangium.
Dictyuchus missouriensis Couch.
Fig. 2. Oogonium.
Figs. 3-5. Stages in germination.
Fig. 6. Germ tube passing through the oogonial wall to the outside.
Fig. 7. Germinated egg with long germ tube and apical sporangium.
Brevilegnia linearis Coker and Braxton.
(Magnification 1816 \times unless otherwise indicated.)
Fig. 8. Oogonium.
Figs. 9-12. Stages in germination.
Fig. 13. Germ tube passing through oogonial wall.
Fig. 14. Tube growing in length.
Fig. 15. Germinated egg with a single long unbranched hypha.
Fig. 16. Another germinated egg which has produced a short, branched germ tube.
Geolegnia inflata Coker and Harvey.
(Magnification 1816 \times unless otherwise indicated.)
Fig. 17. Oogonium.
Figs. 18-20. Stages in germination.
Fig. 21. Germ tube passing from oogonium.
Figs. 22-23. Two examples of germinated eggs. Note long unbranched germ tube. 178 \times .

PLATE 1

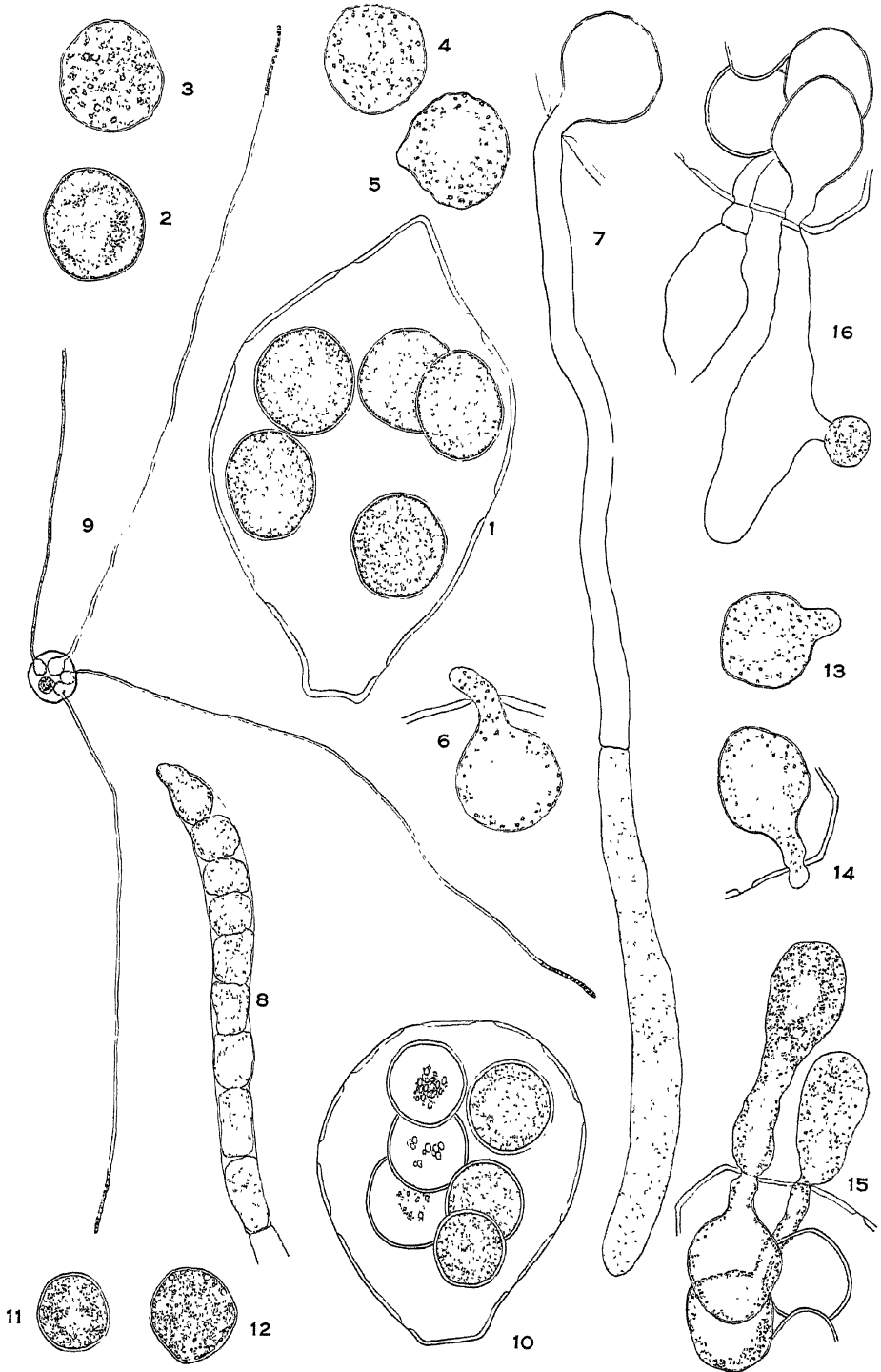


PLATE 2

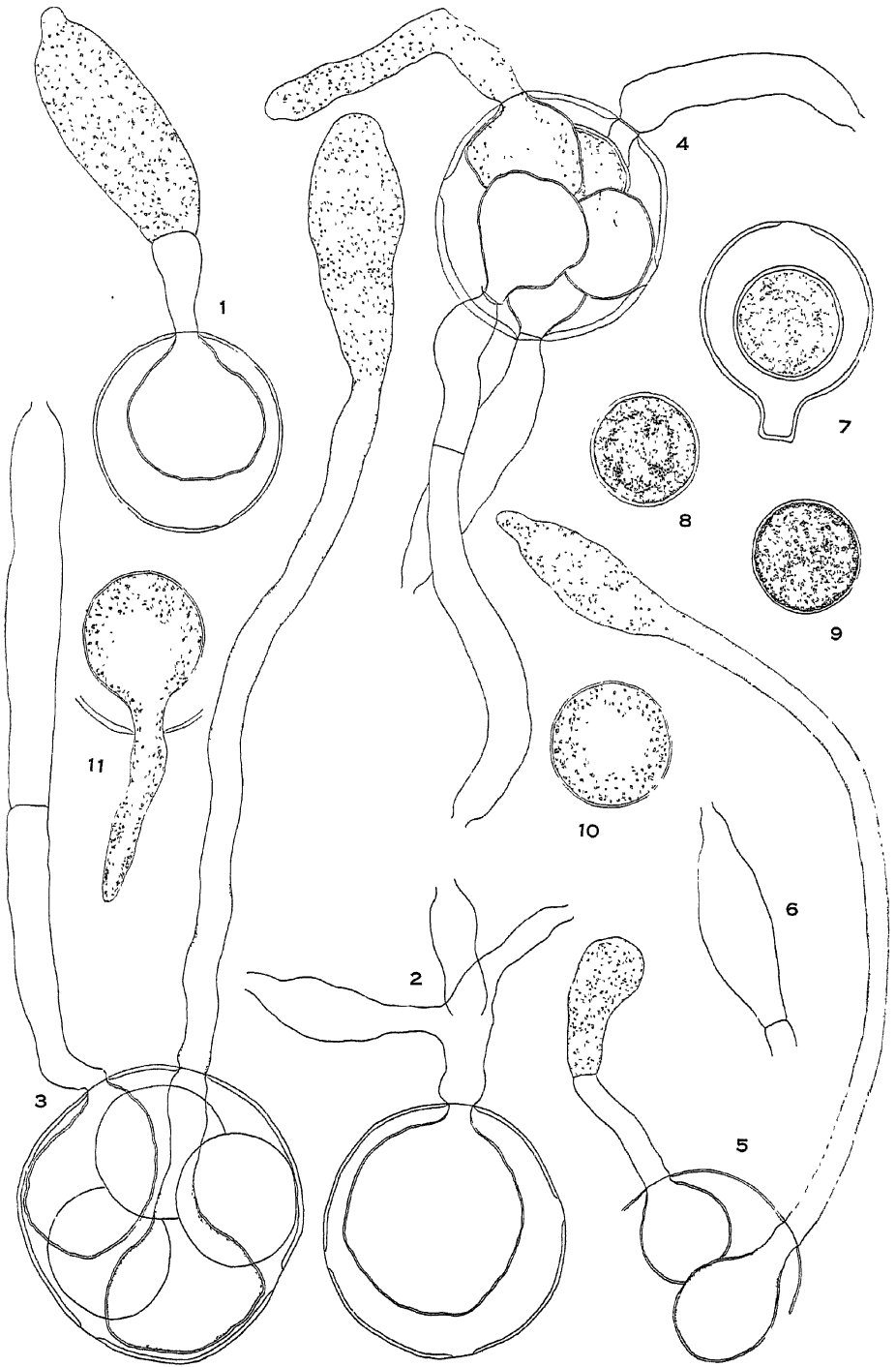


PLATE 3

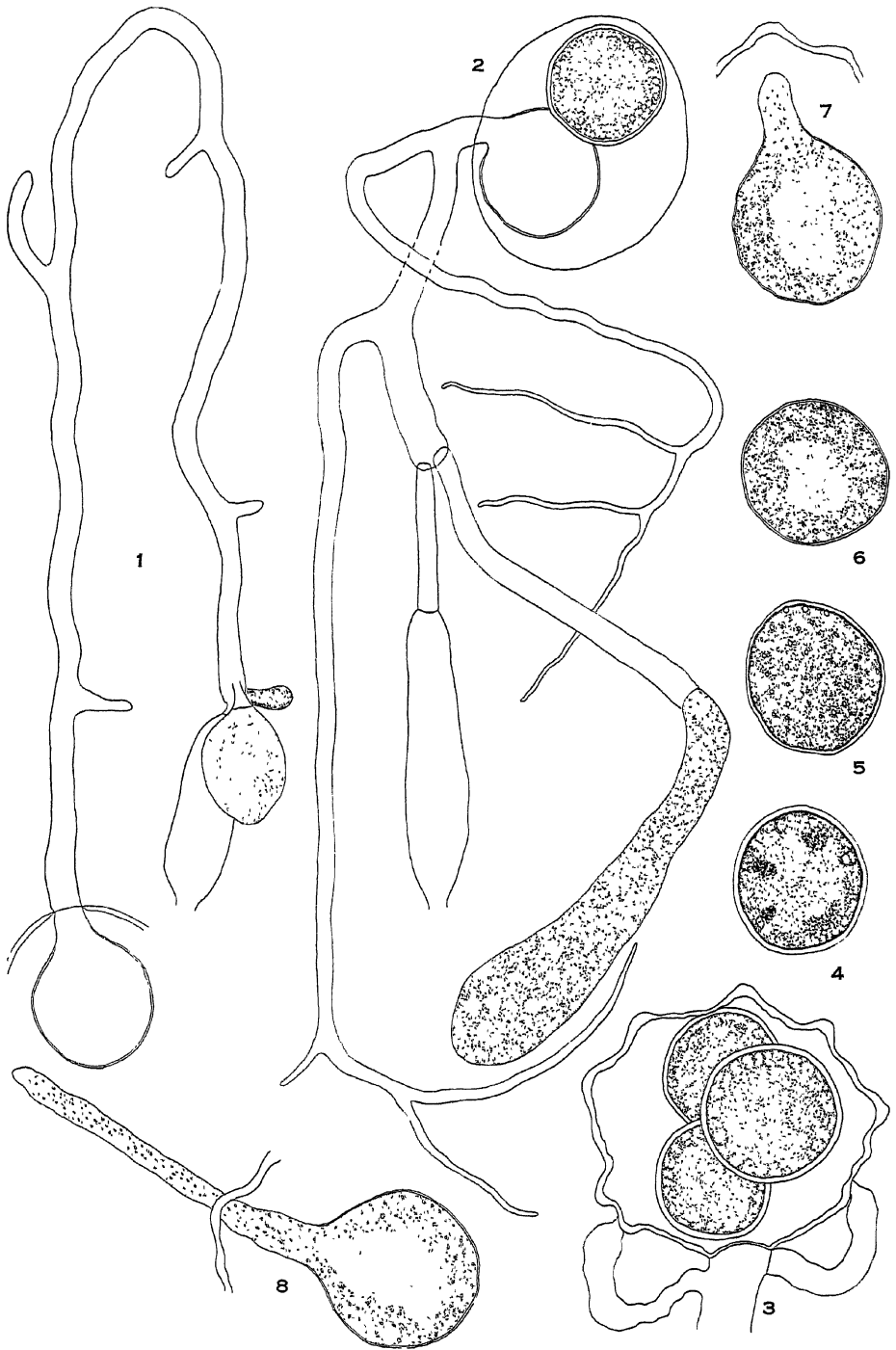


PLATE 4

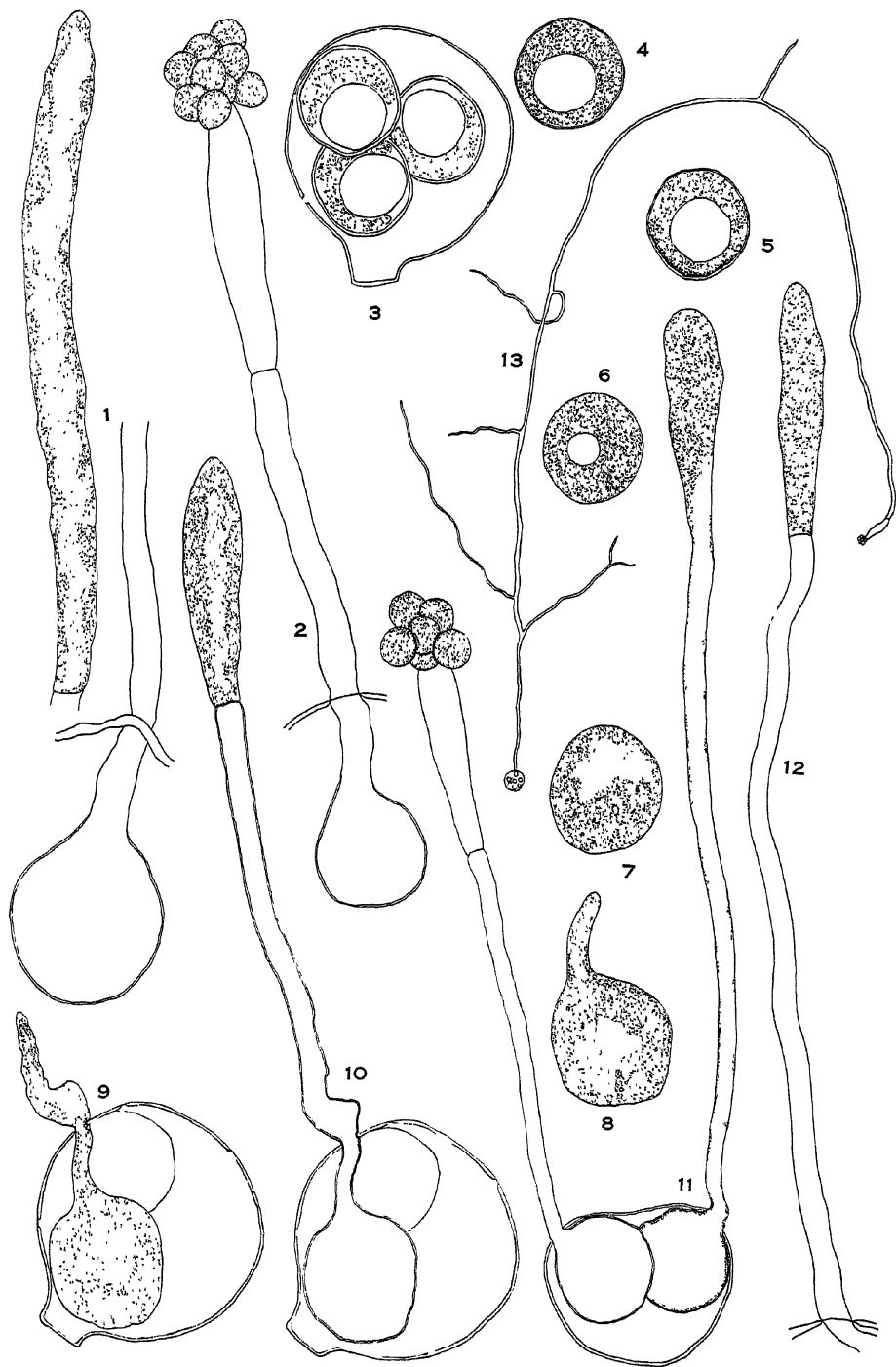


PLATE 5

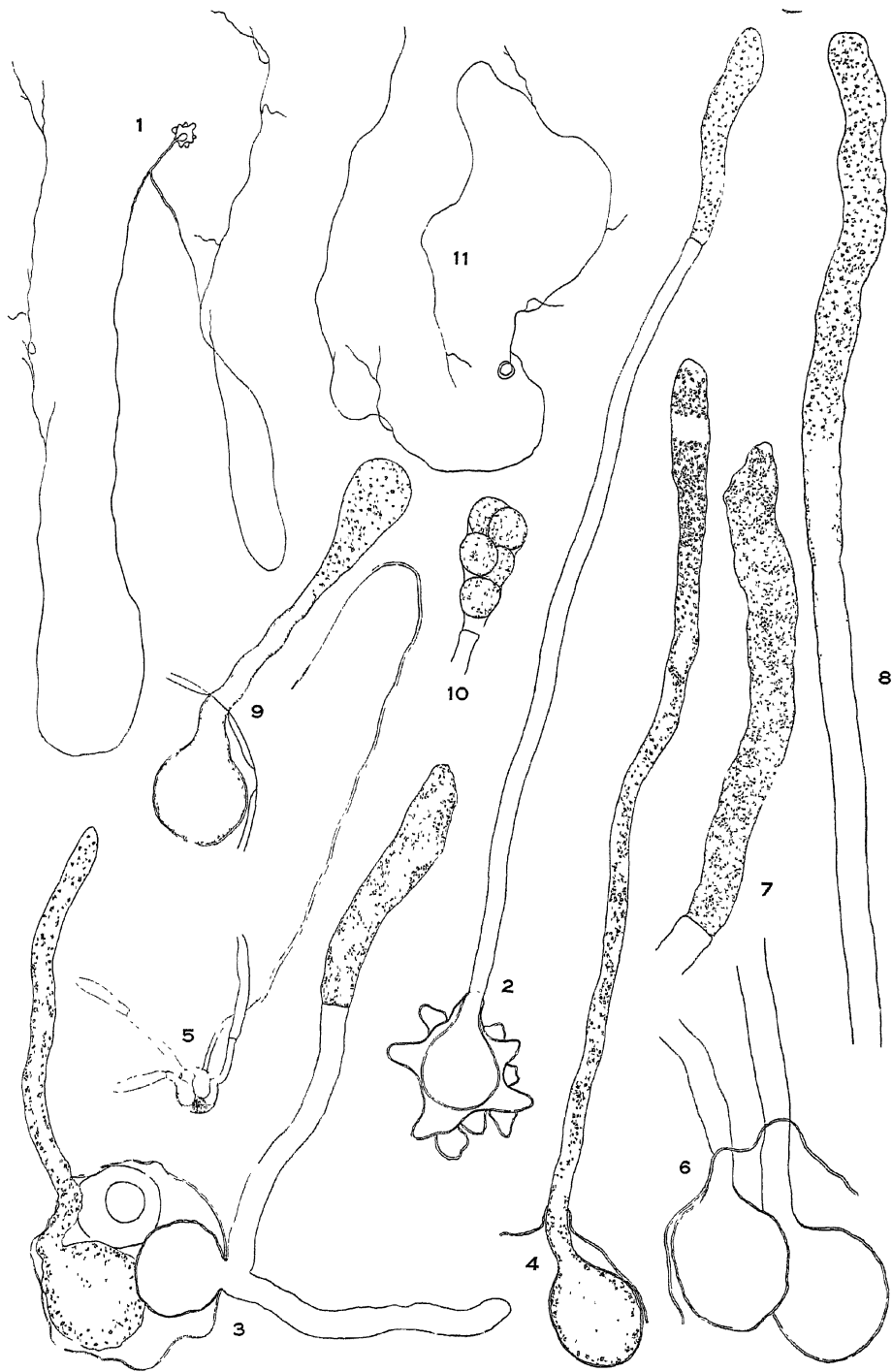
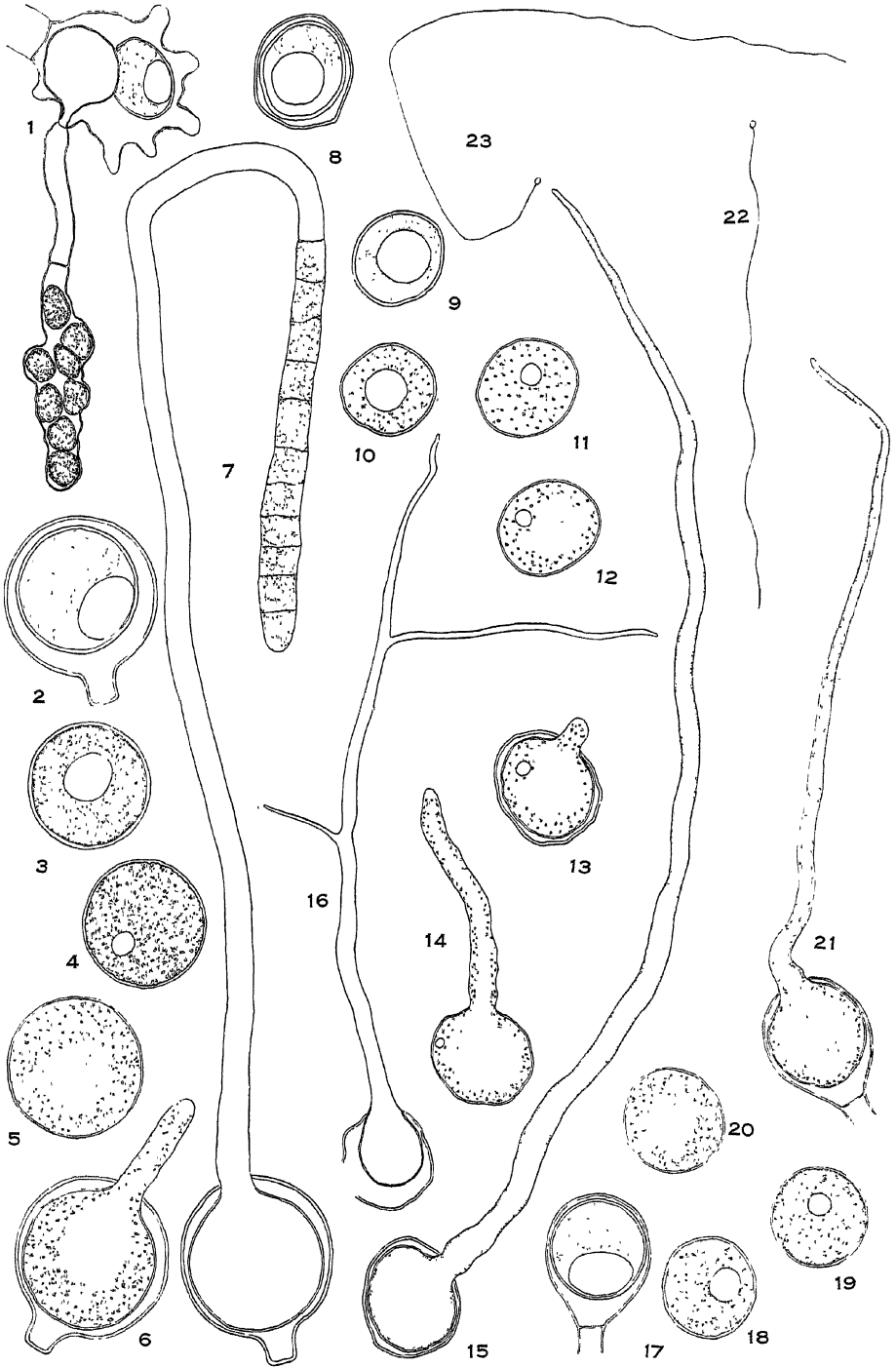


PLATE 6



THE BASKING SHARK, *CETORHINUS MAXIMUS*, ON THE NORTH CAROLINA COAST

By E. W. GUDGER

American Museum of Natural History
New York City

PLATE 7

In 1935, the late H. H. Brimley published in the Mitchell Journal the first records of the occurrence on the North Carolina coast of the two largest sharks that are found in the Atlantic or in any other ocean. One was the Whale Shark, *Rhineodon typus*, the largest of all sharks. This shark, in contrast to the Basking Shark, is very large in the head and shoulders region, has a broad flat head with a terminal mouth beset with many short recurved teeth, and with relatively small or at any rate moderately sized gill slits. The broad head is covered with small and irregularly placed white spots but on the sides of the body the spots are large and set in checker-board squares. The Whale Shark is an inhabitant of tropical and warm-temperate seas. This specimen had drifted north with the Gulf Stream and had become stranded June 6, 1934, on a shoal in Southport Harbor, and at that time was the most northerly known specimen. It was not measured but was estimated to be 40 feet long.

The other big shark recorded by Brimley was the Basking Shark, *Cetorhinus maximus*. This large fish is an inhabitant of cold oceanic waters. In the north Atlantic it is found off the western and northern coasts of Ireland and Scotland, off the eastern coast of Scotland and England, off the Western coast of Norway, and around Iceland. In the western North Atlantic, it comes south in the cold water between the Gulf Stream and the American coast. It was formerly abundant in New England waters (especially in the Gulf of Maine). Occasional specimens are taken off Long Island and along the New Jersey coast. There seems to be no record of its capture in Maryland or Virginia waters, but three records (one old and two new) are now at hand for its occurrence on the North Carolina coast.

Cetorhinus, grayish-brown or slaty in color, is a much slenderer shark than *Rhineodon*. It has a pointed head (like the pointed nose of a bullet in a long cartridge) and a ventral mouth, wide for a shark with a pointed nose. The numerous teeth are small and recurved. *Cetorhinus* is particularly notable for its enormously long gill slits, which almost surround the neck. They begin well up on the dorsum, and those of one side nearly meet on the ventral surface those of the other side. The tail is lunate, the lower lobe being somewhat smaller than the upper. That this shark and its distinctive characters may at this point be made clear to the reader, Figure 1 is introduced here. Its source will be explained later. Like the Whale Shark, the Basking Shark grows to at least 45 feet and numerous specimens of 28 to 35 feet have been measured in New England waters. From this it is clear that the fish is not inaptly named *maximus*. As

will now be seen, our three specimens are relatively small and hence are young fish.

THREE BASKING SHARKS IN OUR COASTAL WATERS

Basking Shark No. I—H. H. Brimley (1935) wrote thus of this specimen: "On Feb. 12, 1935, a female specimen . . . was taken in a shad net about two miles outside Corncake Inlet, . . . a small channel connecting the Cape Fear River with the ocean a few miles below Fort Fisher" [on the south side of the Cape Fear estuary]. This, so far as the available records go, is the farthest south in the western Atlantic for *Cetorhinus*.

Its measurements were: length 13 feet, 5 inches from tip of snout to tip of tail; greatest girth: 53 inches; caudal fin from tip to tip of lobes, 41 inches; width of mouth, across from angle to angle, 18 inches; weight of liver (mostly oil) 65 pounds; weight of fish estimated at about 600 pounds; recurved teeth about one-sixteenth of an inch long; length of gill slits not given.

In order accurately to locate the places of capture of the two 1947 basking sharks, it will be necessary just here to get a clear idea of the coast line of North Carolina from Cape Lookout west and south. From the base of the Cape, the coast (as Shackleford Banks) trends roughly west to Beaufort Inlet, and from Beaufort Inlet, as Bogue Banks, it swings slightly south of west to the localities to be noted where sharks II and III were captured.

Basking Shark No. II—Notice of the capture of and most of the data for this specimen were sent me by Mr. J. W. Bailey, Jr., of Morehead City, N. C., which is situated on the mainland about opposite Beaufort Inlet.

This big fish was taken April 25, 1947, in the Atlantic about half a mile out from Hoop Pole Woods on Bogue Banks, about 10 miles west of Beaufort Inlet. It was captured by Capt. Leo Gillikin of Morehead City in his sinknet fishing boat. This big and logy shark is not much of a fighter, but by its very size and weight, when entangled in the sinknet, it destroyed about 50 yards of this, entailing considerable loss to the fishermen. Fortunately for science, Capt. Gillikin brought this basking shark into Morehead City, where it was an object of much interest.

It was measured over the curves of the body and found to be 13 feet, 6 inches long. Between perpendiculars it would quite surely measure about 13 feet. No scales were available for weighing, but its weight was estimated at about 1300 pounds. A number of photographs were taken. The best (unfortunately with a "gallery" behind it) in left frontal oblique view shows the great size of the fish. The upper jaw being held up by a hook and a rope passing over an overhead pulley, the photograph (kindly sent me by Mr. Aycock Brown) shows the stubby pointed snout, the large wide and vertical gape of the open mouth, and the enormous gill arches. These it may be remarked are the largest and longest found in any shark.

So far as I have found, there has never been published any portrayal by camera or drawing of the inside of the mouth of *Cetorhinus* showing the huge gill arches.

I had wished to use the photograph referred to, but it would require much work by an artist to remove the "gallery". Fortunately there was procured by the American Museum of Natural History in 1930 a splendid *Cetorhinus* taken on the New Jersey coast. It came to us from the fish freezer at Long Branch frozen as solid as a log. When it had thawed out, I had an extensive series of photographs taken of it. Among them that reproduced herein as Figure 1, which shows the fish in side view; and Figure 2, which shows the interior of the mouth and particularly the huge gill arches. This shark was 14 feet, 3 inches between perpendiculars. Since the basking shark is known to reach a length of 45 feet (more than three times longer than the specimen whose mouth is shown in Figure 2) one can only imagine how gigantic its gill arches would be.

Mr. Bailey took some small photographs of this No. II specimen and of these enlargements have been made. One is an oblique-lateral view of the great fish lying on the wharf on its left side. Another is of the pelvic fins and the cloacal region. However, this latter lacks sharpness and contrast, and while it shows what looks very like a clasper, it leaves the sex a bit in doubt. But Mr. Bailey has sent me a sketch of the pelvic fins and the claspers as he saw them. This effectually settles the question that this shark was a male.

Basking Shark No. III—This fine specimen was called to my attention by Mr. Aycock Brown, formerly of Beaufort, but now of New Bern, N. C. It was taken April 25, 1947, by Capt. Gordon Lewis, a sinknet fisherman of Morehead City, in 40 feet of water about 2 miles off Salter Path on Bogue Banks, some 12 miles west of Beaufort Inlet. This shark in its struggles wrapped the lines of the net so tightly around it that it was dead when the fishermen got it out of the net. It is said to have measured 13 feet in length and is estimated to have weighed about 1300 pounds. This estimate is probably about double the actual weight.

Fortunately for this faunal record of the appearance of this fish in the coastal waters of North Carolina, Captain Lewis, with good judgment, brought his big fish to Morehead City, even though this involved towing this 13-foot shark, tail foremost, about 20 miles. What this means I well know, since at Key West years ago we had the "prodigious" task (as we thought) of towing 11-foot tiger sharks 4 miles from the place of capture to the dock where they could be measured and dissected. In Morehead City basking shark No. III was hung up by the tail and photographed. For a copy of the photograph I am indebted to Mr. Ottis Purifoy of Morehead City. This photograph, however, leaves the sex of the fish in doubt.

I had planned to reproduce this photograph herein to show the general form and make up of the shark, but the head is barely clear of the ground and below snout and mouth parts is a heavy shadow. Furthermore, due to the partial eversion of gullet and stomach after the shark had been hanging for some time, the lower mouth parts show as abnormal. These artifacts spoil the photograph for scientific purposes and to my regret it cannot be utilized. However, Figure 1 is made from a photograph which shows the make up of *Cetorhinus* in all points.

Cetorhinus maximus is called basking shark because of its habit of lying awash

at the surface of the sea with its big triangular dorsal fin standing high out of the water. It seems to be literally basking in the sunshine. Being gregarious in its habits, it is said to loaf often in the sunshine in small companies. Several writers have noted that two or three will sometimes swim in tandem fashion with their dorsal fins high out of the water—each observed occurrence possibly giving rise to another sea serpent story.

Possibly in this gregariousness is found the explanation that our basking sharks II and III were taken in the ocean about 2.5 miles apart off the western end of Bogue Banks, on April 25, 1947.

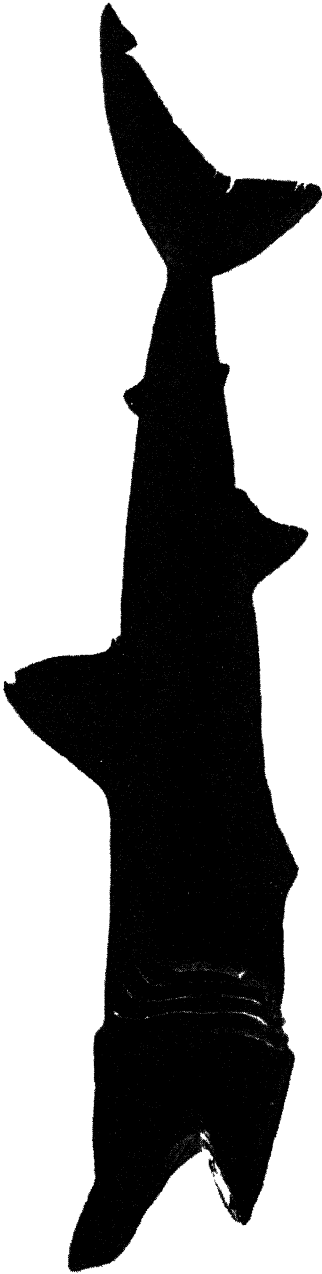
There are rumors of three other large sharks taken in the Cape-Lookout and Shackleford-Banks region this summer, and called basking sharks. But of these Mr. Bailey has not been able to get any definite descriptions, much less any photographs. And because of the newspaper notoriety of the capture of the two sharks referred to herein, it is probable that any large shark taken in this region this summer would be mistakenly called by the fishermen generally—"basking sharks." Hence, in the absence of proof, no notice can be taken of these three alleged sharks of that name.

The basking shark is valuable to shark fishery companies. The thick skin makes good leather, but is so thick that it has to be "split" to make it usable. Thus every skin makes two "sides" of leather for the trade. The meat is ground up for fertilizer. The liver is, as Brimley noted, heavily laden with oil. This when properly purified is used for treating leather for flexibility after tanning. I have somewhere read that years ago there was a basking shark fishery for the liver oil on the western coasts of Ecuador and Peru and a recent newspaper clipping states that at the present day an extensive fishery for *Cetorhinus* is carried on off the southern California coast.

PLATE 7

- Figure 1. The Basking Shark, *Cetorhinus maximus*, in lateral aspect. Note the pointed head, the ventral mouth, the enormously long gill slits and flaps, the high dorsal fin, and the lunate tail fin with upper lobe the longer. Photograph A. M. N. H.
- Figure 2. The Widely Opened Mouth of the Basking Shark. Note the huge gill arches, upper and lower, right and left. Contrast them with the gill slits seen in Figure 1. Note also the pointed snout and in the front upper mouth the oral breathing valve. Photograph A.M.N.H.

PLATE 7



1



2

THE VASCULAR FLORA OF THE OLIVINE DEPOSITS OF NORTH CAROLINA AND GEORGIA*

BY ALBERT E. RADFORD

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina*

PLATE 8 AND THREE TEXT FIGURES

INTRODUCTION

A series of olivine and serpentine deposits are found in the Appalachian mountains from Alabama to Quebec. In North Carolina and Georgia there are 25 high-grade olivine deposits (Hunter, 1941). They are composed of nearly pure magnesium silicates. The high concentration of magnesium in the deposits and the apparent lack of other elements necessary for plant growth suggested that a study of the vegetation on the deposits might prove worthwhile.

No botanical work has been done previously on olivine. Several papers have been published on the flora of serpentine, which is a hydrated magnesium silicate. Schimper (1903) mentioned a few species of ferns characteristic of serpentine in Germany. Harshberger (1903) made an ecological study of the serpentine barrens of Pennsylvania. He found moisture relations the controlling factors in association and species distribution. Fernald (1907), in studying the soil preferences of alpine plants, found 21 vascular species on Mt. Albert, a serpentine mountain in Gaspé County, Quebec; these 21 species are not found elsewhere south of the St. Lawrence River. He also found that species characteristically growing on potassic rocks were dwarfed or stunted when growing on serpentine. However, the more distinctive plants on serpentine were luxuriant wherever there was sufficient moisture. Pennell (1910 and 1912), in a rather extensive study of the flora of the Conowingo Barrens (serpentine) of southeastern Pennsylvania, found 217 species characteristic of the serpentine, and 77 others occurring occasionally. Of those 217 species 17 were restricted to the Conowingo Barrens. Mason (1946) published his study of endemism on serpentine in California. At present, students at the University of California are working on the physiology of several species and the genetics of genera showing great speciation on serpentine.

The objectives of my investigation were to make a taxonomic study of the vascular flora of olivine deposits occurring in North Carolina and Georgia, and to correlate the species and community distribution with the mineral distribution. Nine high-grade olivine deposits in North Carolina and one in Georgia were selected for this study. Every community on each deposit was mapped and at least one or more specimens of every vascular species in each community was

* A thesis submitted to the Faculty of the University of North Carolina in partial fulfillment of the requirements for the degree of Doctor of Philosophy in the Department of Botany.

collected for study. In order to compare the vegetation of the olivine deposits with that of the surrounding areas, two communities on soil derived from rock surrounding the olivine were studied in detail. Observations were made on the flora around the other deposits.

Field studies and collections were made on the ten deposits on the following dates: June 14 to July 13, 1946; August 23-26, 1946; September 6-7, 1946; October 18-20, 1946; May 28 to June 6, 1947; and August 18-23, 1947. Also during August 1947, observations were made on twelve other high-grade olivine deposits in North Carolina and Georgia.

LOCATION OF DEPOSITS

According to Pratt and Lewis (1905), a belt of basic magnesian rocks extends from east-central Alabama, 175 miles from the Gulf of Mexico, to the Gaspé

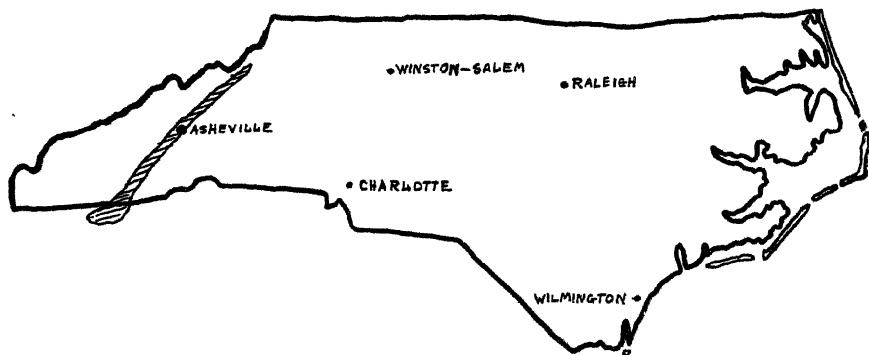


FIG. 1. Outline map of North Carolina showing approximate location of olivine belt.

Peninsula on the St. Lawrence River in Canada, a distance of more than 1600 miles. In North Carolina and Georgia the belt of olivine (basic magnesian rock) is approximately 15 miles wide and 175 miles long, with its widest part at the southern end in Georgia. It extends from Watauga County in North Carolina to White County in Georgia. The center of this belt is near Asheville, North Carolina (Fig. 1).

In 1941 Broadhurst (in Hunter) mapped 25 high-grade olivine deposits for the Tennessee Valley Authority. Of these 25 deposits I selected the following ten (Fig. 2) for botanical study:

- Cane Creek— $3\frac{1}{2}$ mi. SE. of Sylva, Jackson Co., N. C.
- Corundum Hill— $1\frac{1}{4}$ mi. NW. of Gneiss, Macon Co., N. C.
- Dark Ridge— $\frac{1}{2}$ mi. SW. of Balsam P. O., Jackson Co., N. C.
- Day Book—3 mi. N. of Burnsville, Yancey Co., N. C.
- Deposit No. 9— $\frac{1}{2}$ mi. SW. of Ellijay P. O., Macon Co., N. C.
- Frank—Frank, Avery Co., N. C.
- Holcombe Branch— $1\frac{1}{2}$ mi. N. of Democrat, Madison Co., N. C.
- Laurel Creek— $1\frac{1}{2}$ mi. E. of Pine Mt., Rabun Co., Ga.
- Newdale— $1\frac{3}{4}$ mi. E. of Micaville, Yancey Co., N. C.

Newfound Gap—5 mi. NE. of the railroad station at Canton, Haywood-Buncombe Co. line, N. C.

SHAPE AND SIZE OF DEPOSITS

The deposits studied range in width from 250 to 1500 feet, and in length from 1000 to 3000 feet. The largest deposit is Holcombe Branch with an area of .202 square mile (129.28 acres), and the smallest is Newfound Gap with an area of .01 square mile (6.4 acres). The total area of the ten deposits is .453 square mile (290 acres). The areal extent of the deposits was determined by a grid system. The surface areas of the deposits form very irregular patterns (Fig. 3).

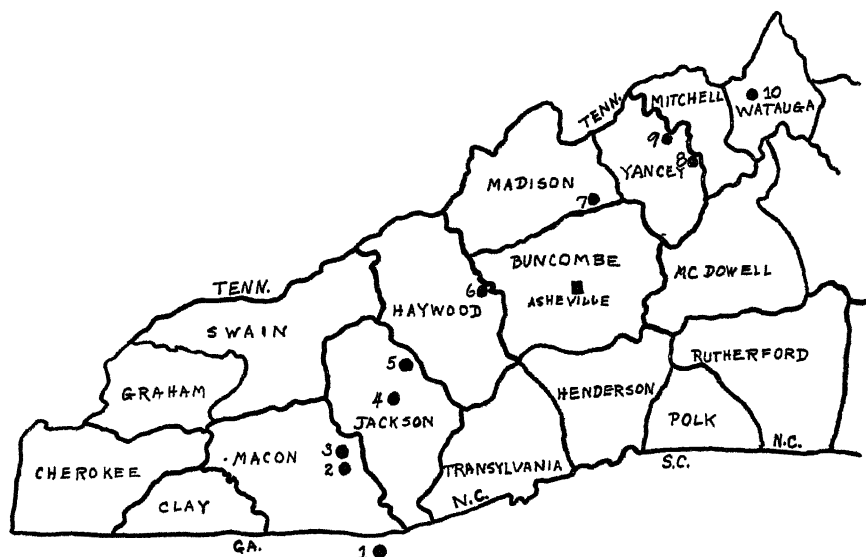


FIG. 2. Counties of western North Carolina showing locations of deposits studied.

GEOLOGY

Regional Geology

The olivine deposits of western North Carolina and northern Georgia are co-extensive with the crystalline rocks of the Appalachian Mountain System. The olivine and other basic magnesian rocks occur in a gneissic belt averaging 35 miles in width, bordered on the east and west by partially metamorphosed sedimentary rocks (Pratt and Lewis, 1905). These basic magnesian rocks form disconnected masses arranged in a line or series of parallel lines that coincide approximately in direction with the strike of the gneissic lamination, N 30° E. Most of the deposits have a sheath of schistose rocks separating them from the normal gneiss.

Hunter in 1941 stated that the deposits are thought to be intrusions that were part of an active pre-Cambrian volcanic system. Olivine deposits now exposed were probably crystallized and cooled at a depth greater than 10 miles below the

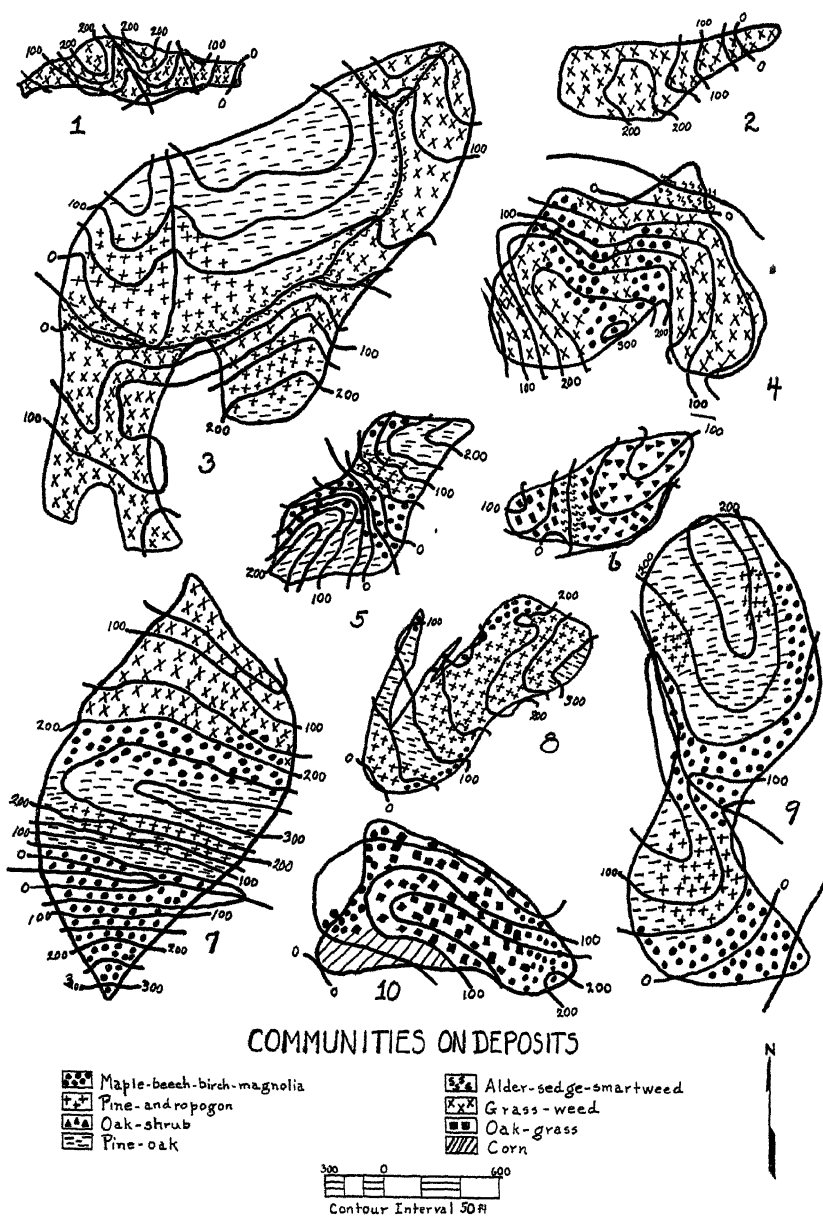


FIG. 3. 1, Cane Creek; 2, Newfound Gap; 3, Holcombe Branch; 4, Frank; 5, Day Book; 6, Newdale; 7, Dark Ridge; 8, Corundum Hill; 9, Laurel Creek; 10, No. 9.

surface at the time of their injection. During geological times, rocks and sediments to a depth of approximately 10 miles have been removed from this area by erosion.

Petrology

The deposits selected for study are composed of dunite and saxonite which are essentially pure olivine. Chromite is the primary accessory mineral of dunite, and bronzite is the primary accessory mineral of saxonite.

These deposits can be divided into two structural types, the ring dike, and the lens-shaped with its long axis parallel to the regional schistosity. Cooling joints and occasional faults are prominent features of the lens-shaped structure. The most noticeable feature of the ring dike is the parallel lamination which on a weathered surface has the appearance of a sandstone. Veins of different minerals are present in each deposit.

Chemical analyses of dunite and saxonite are given in Table II.

TABLE I
Size of Deposits

DEPOSIT	MAX. LENGTH	MAX. WIDTH	SQ. MI.	ACRES
Cane Creek	1500 ft.	350 ft.	0.016	10.24
Corundum Hill	1200 ft.	500 ft.	0.017	10.88
Dark Ridge	2000 ft.	900 ft.	0.047	30.08
Day Book	2000 ft.	600 ft.	0.035	22.40
Deposit No. 9	1000 ft.	800 ft.	0.02	12.80
Frank	1400 ft.	600 ft.	0.031	19.84
Holcombe Branch	3000 ft.	1500 ft.	0.202	129.28
Laurel Creek	2500 ft.	800 ft.	0.045	28.80
Newdale	1800 ft.	500 ft.	0.03	19.20
Newfound Gap	1000 ft.	250 ft.	0.01	6.40

Mineralogy

Olivine usually accounts for more than 60% of the minerals found in dunite and saxonite. Most of the olivine consists of 80% forsterite (Mg_2SiO_4), 11% or less fayalite (Fe_2SiO_4), and the remaining 9% or less of other magnesian minerals which are usually hydrates derived from olivine.

The high-grade olivine, 45% or more MgO, is usually found in the center of the deposits. The area on the deposits surrounding this high-grade olivine is composed mainly of faulted and partly serpentinized dunite and saxonite. Between the surrounding gneisses and schists, and the serpentinized dunite and saxonite, is a talcy vermiculite fringe zone a few feet in width (Hunter, 1941).

Listed below are the secondary magnesian and accessory minerals found on the deposits studied:

Anthophyllite	$(\text{MgFe})_7\text{Si}_8\text{O}_{22}(\text{OH})_8$
Bronzite	$(\text{MgFe})\text{SiO}_3$
Chlorite	$(\text{MgFe})_3(\text{AlFe})_2\text{Si}_5\text{O}_{16}(\text{OH})_8$
Chromite	FeCr_2O_4
Serpentine	$\text{Mg}_3\text{Si}_2\text{O}_5(\text{OH})_4$
Talc	$\text{Mg}_3\text{Si}_4\text{O}_{10}(\text{OH})_2$
Vermiculite	$(\text{OH})_2(\text{MgFe}_3)(\text{Si}, \text{Al}, \text{Fe})_4\text{O}_{10} \cdot 4\text{H}_2\text{O}$

(Minerals from Hunter, 1941; chemical formulae from Hurlbut, 1941).

TOPOGRAPHY

Most of the deposits studied occur as resistant ridges or as exposed outcrops on steep hillsides. Small streams have cut through the Dark Ridge, Day Book, and Laurel Creek deposits, forming olivine cliffs on either side, some of which rise as high as 375 feet above the stream level. The North Toe River cuts through the Frank Deposit. The deposits range in elevation from 1800 feet on Laurel Creek, lowest point on the deposit, to approximately 3250 feet on Newfound Gap.

SOILS

The deposits are characterized by numerous small boulders, and by depressions that are formed by chemical weathering. Residual clays occur in the joints and

TABLE II
Chemical Analysis of Rocks
Compiled from Hunter, 1941

ROCK	DEPOSIT	MgO	SiO ₂	Fe ₂ O ₃	Ti, Cr, Al (OXIDES)	CaO	ION. LOSS	ALK.
Fine-grained Dunite (unaltered Olivine)	Day Book	49.31	40.86	7.66	2.18	0.00	0.63	
Fine-grained Saxonite (partly altered Olivine).	Day Book	45.92	42.40	8.62	1.06	0.00	1.23	
Coarse-grained Dunite (unaltered Olivine)	Dark Ridge	49.69	41.00	9.74	0.38	0.00	0.61	
Weathered Dunite	Dark Ridge	44.08	43.60	9.90	1.76	1.23	1.51	
Serpentinized Saxonite	Cane Creek	44.82	41.34	9.44	1.58	0.30	2.35	
Dunite: unaltered Olivine	Cane Creek	48.78	40.04	10.24	0.48	0.26	0.92	
Dunite: unaltered Olivine	No. 9	47.50	38.94	10.88	1.36	0.08	0.89	
Dunite: unaltered Olivine	Corundum Hill	49.35	40.58	8.30	0.65	trace	1.13	
Partly Serpentinized Dunite	Corundum Hill	45.46	41.60	8.46	1.70	trace	2.61	
Serpentinized Dunite	Laurel Creek	42.17	43.92	8.94	1.40	0.00	3.94	
Dunite (average of five commercial shipments)	Day Book	48.77	40.93	7.60	1.32	0.29	1.09	0.13

depressions. These reddish and yellowish clays are seldom over six inches deep except in a few joints or near the borders of deposits on slopes. There is no mature soil on any of the deposits. The more nearly level the deposit, the deeper are the clays. The streams crossing the deposits are rapidly down-cutting, thus very little transported material is found along the banks.

The deposits occur as ridges or outcrops. The olivine is more resistant than the surrounding rocks. It weathers slowly by chemical decomposition. Where seams or veins of vermiculite or anthophyllite occur in the deposit, mechanical weathering takes place. Deposit No. 9 supports a more mesic vegetation on the south slope because the physical weathering of anthophyllite and vermiculite has produced a deeper and better moisture-holding soil. In general, however, erosion on the slopes and rapid leaching of the soluble compounds prevent the accumulation of soil materials.

Very little is known about the formation of soil from olivine, but Merrill (1921) states: "The deep green serpentines of Harford County, Maryland, weather slowly down into a gray-brown soil, which consists of 60.17% silica, 10.40% of the iron oxides, 14.81% of alumina, and only 7.23% magnesia. A fresh rock, on the other hand, carries nearly 40% of magnesia, 8.50% iron and other metallic oxides, and less than one-half of one per cent of alumina."

Ten samples of residual clays from communities on various deposits were tested for acidity and alkalinity. The Beckman pH meter was used in making the tests. 10 grams of air-dried clay were mixed with 25 cc. of distilled water and left overnight in a closed container before testing. The samples taken from the surface clays to a depth of one or two inches were acid, the average pH being 6.3. Two samples taken from the bottom inch above the parent rock were slightly alkaline, pH 7.6 and 7.4.

Judging from the distribution of ericaceous plants, which grow only where the residual clays are 4 or more inches in depth, and from the results of the pH tests, it seems that the surface layers of the deeper clays are acid. Apparently a high percentage of the Mg ions are leached as soon as the olivine goes into solution.

Gordon and Lipman (1930), in a study on why soils derived from basic magnesian rocks in California are infertile, found that the magnesium had no toxic effect on the plants. They concluded that the high pH (about 8.1), and nitrate and phosphate deficiencies were responsible for poor growth. Dr. Lutz, Department of Agriculture, Soils Division, Raleigh, N. C., in unpublished observations said that soils derived from dunite and serpentine are some of our most productive in North Carolina when lime, nitrates, and phosphates are added as fertilizers. A farmer reported to me that his corn yield on Deposit No. 9 was greater than that of a similar acreage of bottom land. He used lime as a fertilizer and planted near the base of the deposit where the clays were six inches deep or deeper. The natural vegetative growth on the deposits is luxuriant where there is abundant moisture.

CLIMATOLOGY

The climate of the mountain region of western North Carolina is a modified continental type. The summers are cooler and the winters are milder than corresponding seasons in the interior states. Valleys and ridges and differences in elevation are responsible for sharp climatic variations in relatively small areas.

The rainfall ranges from 39.13 inches annually at Marshall in the central mountain region, to 82.63 inches at Highlands in the southern mountain region. The January temperature average is approximately 38°F. and the July average is approximately 71°F. for the entire region. The first killing frosts in the fall usually occur the second week of October, and the last killing frost of spring occurs the last week of April.

Although data from weather stations in the mountain region show climatic differences, the vegetation on the deposits appears to be essentially the same throughout the olivine belt.

PLANT COMMUNITIES

General

All of the communities on olivine are characterized by a great number of species but by very few individuals of any single species. I have described seven communities on the deposits and one community on soils derived from rocks surrounding the olivine. These communities are: (1) maple-beech-birch-magnolia; (2) pine-andropogon (grass); (3) pine-oak; (4) oak-grass; (5) oak-shrub; (6) grass-weed (pasture on dunite); (7) alder-sedge-smartweed; and (8) grass-weed-oak (pasture on soils derived from rocks surrounding the olivine deposits).

With the exception of the pine-andropogon (various grasses and small herbs) community which is the most striking one on the deposits, the others are more or less typical of the mountain region. I have not observed a pine (pitch and scrub)-andropogon community above 2000 feet in the Blue Ridge except where pines have been planted for soil conservation purposes. The only large pine communities that I have observed are those composed of white pines in the wetter locations of the mountains.

On the north slopes of the deposits where moisture is more abundant, a maple-beech-birch-magnolia community is found. On the drier south slopes with other edaphic conditions the same as those on the north slopes, a pine-andropogon community is found. If there is more soil material, therefore more moisture, on a south slope, a pine-oak or oak-shrub community is found.

My observations on the controlling factors in distribution of species and communities agree in general with those of Harshberger (1903), who studied the serpentine barrens of Pennsylvania. He states that "one fact is proved abundantly by a study of the flora of the serpentine barrens and that is that the chemical character of the soil derived from the disintegration of serpentine plays an unimportant part in the distribution of the plants mentioned. The distribution of such species is due rather to the physical conditions of the soil especially with reference to water conductivity and water storage conditions. The variation and character of the plant associations described above is in main due to the character of the soil. If the soil is present as a well marked surface layer then tree associations are found, if on the other hand the rock is exposed, herbaceous associations are the rule. The surface layers of serpentine rock are broken by weathering into angular fragments, which lying loosely together, permit the percolation of the rain water down into the seams of the underlying rock. Such exposures therefore, support plants that have adapted themselves to living in dry situations and have structural arrangements which prevent a rapid loss of water." Soil acidity or alkalinity is also a very important factor in species distribution.

All of the deposits have been cut over in the last 60 years and several have been mined. Cane Creek, Holcombe Branch, Frank, and Newfound Gap Deposits are partly seeded pastures at present. Cultivation has left its imprint on the community pattern on Deposit No. 9. The maple-beech-birch-magnolia community on the Day Book Deposit has been destroyed during the past year by highway construction.

The communities found on each of the deposits are as follows: Cane Creek, grass-weed; Corundum Hill, maple-beech-birch-magnolia, pine-andropogon, pine-oak; Day Book, maple-beech-birch-magnolia, pine-andropogon, pine-oak; Dark Ridge, maple-beech-birch-magnolia, pine-andropogon, pine-oak, grass-weed; No. 9, maple-beech-birch-magnolia, oak-grass; Frank, grass-weed, maple-beech-birch-magnolia, alder-sedge-smartweed; Holcombe Branch, pine-andropogon, grass-weed, pine-oak, alder-sedge-smartweed; Laurel Creek, maple-beech-birch-magnolia, pine-oak, pine-andropogon; Newfound Gap, grass-weed.

TABLE III
pH Tests on Residual Clays

COMMUNITY	DEPOSIT	pH
Maple-beech-birch-magnolia	Day Book	7.0
Maple-beech-birch-magnolia	Dark Ridge	6.3
Maple-beech-birch-magnolia	Laurel Creek	7.6
Pine-oak	Holcombe Branch	5.2
Pine-oak	Dark Ridge	5.4
Pine-andropogon	Laurel Creek	6.8
Pine-andropogon	Holcombe Branch	6.65
Pine-andropogon	Day Book	7.4
Grass-weed	Holcombe Branch	6.5
Oak-grass	Deposit No. 9	6.8

Following are the abbreviations of the deposits and the communities, and their meanings as employed in the following pages:

<i>Deposits</i>	<i>Communities</i>
CC—Cane Creek	mbm—maple-beech-birch-magnolia
CH—Corundum Hill	og—oak-grass
DB—Day Book	os—oak-shrub
DR—Dark Ridge	pa—pine-andropogon (grass)
D9—No. 9	pas—grass-weed-oak (pasture on surrounding rock or soils derived therefrom)
F—Frank	
HB—Holcombe Branch	pd—grass-weed (pasture on dunite)
LC—Laurel Creek	po—pine-oak
N—Newdale	s—alder-sedge-smartweed
NG—Newfound Gap	

Description of Communities

1) *Maple-beech-birch-magnolia community*

This community which is highly mesophytic is characterized in general by dense vegetation and is particularly well-developed on the northern slopes near the streams. It occurs on the Corundum Hill, Dark Ridge, Day Book, No. 9, Frank, and Laurel Creek Deposits.

On the Corundum Hill Deposit *Fagus grandifolia*, *Acer rubrum*, and *Betula lenta* are the dominant trees. These trees are 4–12 inches in diameter and 20–40 ft. tall. *Cornus florida* and various saplings form a scattered irregular

layer beneath the tree cover. The most common shrubs scattered through the community are *Rhododendron calendulaceum*, *Benzoin aestivale*, *Kalmia latifolia*, and *Hydrangea arborescens*. The herbaceous ground cover consists of a wide variety of species, with the ferns *Osmunda cinnamomea*, *Adiantum pedatum*, *Athyrium thelypteroides*, and *Athyrium asplenoides* being the most common. Some of the other more common herbaceous species are *Cimicifuga racemosa*, *Heuchera americana*, *Luzula campestris* var. *bulbosa*, *Sanicula canadensis*, *Glycyrrhiza melicaria*, *Nepeta hederacea*, *Oxalis corniculata*, *Ranunculus recurvatus*, and *Thalictrum dioicum*. *Phlox stolonifera* forms a patch covering nearly 40 sq. ft. near the stream.

On the Dark Ridge Deposit *Acer pennsylvanicum*, *A. rubrum*, *Magnolia acuminata*, and *Betula lenta* are the dominant trees. These are 4-10 inches in diameter, and 25-50 ft. tall. *Calycanthus fertilis*, *C. nanus*, *Evonymus obovatus*, *Pyrularia pubera*, *Hydrangea arborescens*, *Rubus occidentalis*, and *R. odoratus* are the common shrubs. The herbaceous vegetation is the most luxuriant found in any community. *Tiarella cordifolia*, *Thaspium aureum*, *Zizia cordata*, *Disporum lanuginosum*, *Trillium erectum*, *Polystichum acrostichoides*, *Smilacina racemosa*, *Polygonatum biflorum*, *Clintonia umbellulata*, *Arabis canadensis*, *Gentiana quinquefolia*, and *Aster divaricatus* are the more common herbaceous species. *Polypodium virginianum* and several species of mosses form a mat on the olivine boulders. One of these boulders 20 feet above stream level is covered by a patch of *Campthorhynchus rhizophyllus* about 10 sq. ft. in area.

On the Day Book Deposit *Pinus Strobus*, *Tsuga canadensis*, *Acer rubrum*, and *Liriodendron tulipifera* are the dominant trees. These trees are 4-12 inches in diameter and 20-50 ft. tall. *Kalmia latifolia* and *Rhododendron maximum* form a dense entanglement beneath the *Pinus Strobus* and *Tsuga canadensis*. *Alnus rugosa*, *Benzoin aestivale*, *Pyrularia pubera*, *Cornus alternifolia*, and *Sambucus pubens* are the more common shrubs beneath *Acer rubrum* and *Liriodendron tulipifera*. *Carex pennsylvanica*, *C. digitalis*, *Stellaria pubera*, *Polygonatum commutatum*, *Smilacina racemosa*, and *Trillium erectum* are the more common herbaceous species. *Stenanthium gramineum* forms several large colonies. *Phlox stolonifera* also forms one large colony. Mosses, lichens, and *Polypodium virginianum* form thick carpets on the olivine boulders in this community.

On Deposit No. 9 the maple-beech-birch-magnolia community is drier than on the other deposits. *Fagus grandifolia*, *Quercus falcata*, *Q. alba*, and *Q. coccinea* are the dominant trees. These are 4-15 inches in diameter and 20-50 ft. tall. *Cornus florida*, *Sassafras albidum*, and numerous other saplings form an irregular layer beneath the tree cover. The common shrubs are *Vaccinium stamineum*, *Hydrangea arborescens*, and *Corylus americana*. *Adiantum pedatum*, *Carex rosea*, *Circaea latifolia*, *Podophyllum peltatum*, *Sabatia angularis*, *Sanicula canadensis*, and *Scutellaria incana* are the common herbaceous species. *Frasera carolinensis* forms a large colony in the cove at the western end of the deposit.

On the Frank Deposit this community is approximately 100 feet above the stream level. *Acer rubrum*, *Quercus borealis* var. *maxima*, *Betula lenta*, *Fagus grandifolia*, and *Magnolia acuminata* are the dominant trees. These trees are

6–15 inches in diameter and 30–60 ft. tall. *Rhododendron maximum* and *Kalmia latifolia* form dense entanglements in parts of the community. *Clethra acuminata*, *Hydrangea arborescens*, and *Rhododendron calendulaceum* are the common shrubs. *Adiantum pedatum*, *Aster divaricatus*, *Cystopteris fragilis*, *Polystichum acrostichoides*, *Carex varia*, *Silene stellata*, and *Iris cristata* are the common herbs: *Camptosorus rhizophyllus* forms small patches of a square foot on several of the olivine boulders. Mosses and lichens form thin mats or carpets on other boulders in this community.

On the Laurel Creek Deposit *Pinus Strobus* and *Tsuga canadensis* are the dominant trees along the streams, and a little higher on the slopes where there is less moisture, *Acer rubrum* and *Fagus grandifolia* are dominant. These trees are 6–15 inches in diameter and 20–50 ft. tall. *Rhododendron maximum* and *Kalmia latifolia* are the common shrubs beneath *Pinus Strobus* and *Tsuga canadensis*. *Gaylussacia ursina*, *Hydrangea arborescens*, *H. cinerea*, and *Asimina triloba* are the more common shrubs beneath *Acer rubrum* and *Fagus grandifolia*. *Sanicula canadensis*, *Mitchella repens*, *Collinsonia canadensis*, *Tiarella cordifolia*, *Mitella diphylla*, *Thaspium aureum*, and *Asarum grandiflorum* are the common herbaceous species. *Conopholis americana* forms a 40 square foot patch in the central cove.

2) Pine-andropogon community

This xerophytic community is characterized by an open stand of pines which are mostly 20–30 feet apart, with a grass-herbaceous ground cover through which are scattered low shrubs. This type of community is found on the dry southern slopes of the Corundum Hill, Day Book, Holcombe Branch, and Laurel Creek Deposits.

On the Corundum Hill Deposit *Pinus rigida* and *P. virginiana* are the dominant trees. *Quercus falcata* and *Q. stellata* are the sub-dominants. The pines are 9–20 inches in diameter and 30–40 ft. tall. At least one fourth of the pines are forked at the base. The oaks are 2–6 inches in diameter and seldom over 20 ft. tall. Along the mine veins are found *Acer rubrum*, and *Liriodendron tulipifera* saplings which have yellowish leaves and appear dwarfed. *Gaylussacia baccata*, *Vaccinium stamineum*, *Physocarpus opulifolius*, *Kalmia latifolia*, and *Rhododendron maximum* are the characteristic shrubs. *Alnus rugosa* occurs along the mine veins. *Andropogon furcatus*, *Panicum sphaerocarpon*, *Panicum lanuginosum* var. *fasciculatum*, *Gaultheria procumbens*, *Liatris spicata*, *Hypoxis hirsuta*, *Eryngium aquaticum*, *Lespedeza hirta*, *Hieracium venosum*, *Thalictrum revolutum*, *Sisyrinchium angustifolium*, and *S. mucronatum* are the characteristic herbs.

On the Day Book Deposit *Pinus rigida* is the dominant tree. *Quercus alba*, *Q. stellata*, and *Q. borealis* var. *maxima* are sub-dominants. The pines are 6–8 inches in diameter and 30–40 ft. tall; most of the trunks are crooked. The two specimens of *Quercus stellata* are 15 inches in diameter and 30 ft. tall. *Pinus Strobus* is sparsely intermingled with *P. rigida*. *Betula lenta* growing in this community is stunted, while *Magnolia acuminata* seems to be normal. *Ceanothus*

americanus, *Vaccinium stamineum*, *Rhododendron calendulaceum*, *Kalmia latifolia*, and *Rhododendron maximum* are the common shrubs scattered through the community. *Panicum Boscii*, *P. Ashei*, *P. lanuginosum* var. *fasciculatum*, *Andropogon scoparius*, *Solidago nemoralis*, *Thalictrum revolutum*, *Asclepias verticillata*, *Lespedeza hirta*, *Lobelia inflata*, *Anychia polygonoides*, *Desmodium rigidum*, *D. marilandicum* and *Aster patens* are the common herbaceous species.

On the Holcombe Branch Deposit *Pinus rigida* is the dominant tree in the pine-andropogon community. *Quercus falcata* and *Q. stellata* are sub-dominants. These trees are 6–10 inches in diameter and 20–40 ft. tall. *Ceanothus americanus*, *Lyonia ligustrina* var. *foliosiflora*, *Vaccinium stamineum*, and *Kalmia latifolia* are the common shrubs present. *Andropogon scoparius*, *Panicum sphaerocarpon*, *Scleria ciliata*, *Sericocarpus asteroides*, *Solidago nemoralis*, and *Thalictrum revolutum* are the common herbaceous species. Lichens are abundant on the olivine boulders and on the trees in the community.

On the Laurel Creek Deposit *Pinus virginiana* and *Quercus stellata* are the dominants. The pines are 2–8 inches in diameter and 15–25 ft. tall; the oaks are 4–8 inches in diameter and 10–40 ft. tall. *Ceanothus americanus* and *Kalmia latifolia* are the common shrubs. *Andropogon furcatus*, *Panicum commutatum*, *P. lanuginosum* var. *fasciculatum*, *P. dichotomum*, *Stipa avenacea*, *Melica mutica*, *Asclepias verticillata*, *Sericocarpus asteroides*, *Chrysopsis mariana*, *C. graminifolia*, *Liatris graminifolia*, and *Sisyrinchium angustifolium* are the common herbaceous species.

3) Pine-oak community

This is a transition community between the pine-andropogon and the maple-beech-birch-magnolia communities. Pines and oaks predominate, but in general the species composition is mixed. This community occurs on the Dark Ridge, Day Book, Holcombe Branch, and Laurel Creek Deposits.

On the Dark Ridge Deposit, the plants in this community grow mostly in the joints between large olivine boulders. *Pinus rigida*, *P. virginiana*, *Quercus stellata*, *Q. borealis* var. *maxima*, *Q. alba*, and *Q. montana* are the dominant trees. The pines are 6–10 inches in diameter and 20–40 ft. tall. The oaks are 8–12 inches in diameter and 30–50 ft. tall. The shrubs form an almost impenetrable thicket. The shrubs are *Ceanothus americanus*, *Kalmia latifolia*, *Calycanthus fertilis*, *C. nanus*, *Pyrolaria pubera*, *Vaccinium stamineum*, *Rhododendron calendulaceum*, woven together by *Smilax glauca*, *S. rotundifolia* var. *quadrangularis*, *Rubus allegheniensis*, and *R. occidentalis*. In the few clearings in the shrub thicket are *Aletris farinosa*, *Gerardia tenuifolia*, *Panicum lanuginosum* var. *fasciculatum*, *Polygala Senega*, *Phlox carolina*, *Galax aphylla*, *Tradescantia montana* var. *subaspera*, *Euphorbia corollata*, and *Coreopsis major*. Very few lichens are found on the olivine boulders of this community.

On the Day Book Deposit *Pinus Strobus*, *Quercus alba*, *Q. borealis* var. *maxima*, *Q. coccinea*, *Q. falcata*, *Q. stellata*, *Oxydendrum arboreum*, and *Nyssa sylvatica* are the dominant trees. These are 6–18 inches in diameter and 20–70 ft. tall. *Rhododendron maximum*, *Ceanothus americanus*, *Vaccinium stamineum*, *Lyonia*

ligustrina, *Sassafras albidum*, and *Rhododendron calendulaceum* are the common shrubs. The more common herbs are *Chrysopsis mariana*, *Coreopsis major*, *Hypericum punctatum*, *Lespedeza hirta*, *Panicum Boscii* var. *molle*, *Pteridium aquilinum* var. *latiusculum*, *Thalictrum revolutum*, *Euphorbia corollata*, and *Aster patens*. Lichens are abundant on the few exposed olivine boulders of this community.

In the pine-oak community on the Holcombe Branch Deposit, *Pinus rigida*, *Quercus stellata*, *Q. falcata* are the dominant trees. These are 3-20 inches in diameter and 10-60 ft. tall, averaging nearly 30 ft. tall. *Castanea pumila*, *Kalmia latifolia*, *Gaylussacia baccata*, *Ceanothus americanus*, *Vaccinium stamineum*, and *Viburnum prunifolium* are the more common shrubs scattered through the community. The common herbaceous species are *Panicum dichotomum*, *P. Boscii* var. *molle*, *Hypericum punctatum*, *Sericocarpus asteroides*, *Thalictrum revolutum*, *Stylosanthes riparia*, *Pteridium aquilinum* var. *latiusculum*, *Chrysopsis mariana*, *Chimaphila maculata*, *Solidago Boottii*, *Sanicula canadensis*, and *Erigeron canadensis*. A few lichens and mosses are found on the olivine boulders.

On the Laurel Creek Deposit *Pinus virginiana*, *Quercus stellata*, *Q. borealis* var. *maxima*, *Nyssa sylvatica*, and *Acer rubrum* are the dominant trees. They are 6-8 inches in diameter and 15 to 30 ft. tall. Several of the trees are stunted and gnarled. *Ceanothus americanus*, *Vaccinium stamineum*, *Lyonia ligustrina*, *Kalmia latifolia*, *Smilax glauca*, and *Vitis rotundifolia* are the common shrubs. *Vitis rotundifolia* forms an extensive ground cover on the northern slope of the deposit. Ferns and grasses compose a large percentage of the herbaceous species. The more common ferns are *Cheilanthes lanosa*, *Polypodium polypodioides*, *Polystichum acrostichoides*, *Asplenium platyneuron*, *A. Trichomanes*, and *Osmunda regalis* var. *spectabilis*. *Melica mutica*, *Stipa aenacea*, *Danthonia sericea*, *Panicum dichotomum*, *P. depauperatum*, and *P. Boscii* are the common grasses. Other common herbs are *Sericocarpus asteroides*, *Sisyrinchium gramineum*, *S. mucronatum*, *Euphorbia corollata*, *Scleria oligantha*, and *Silene virginica*. Lichens are abundant on the boulders.

4) Oak-grass community

This community is characterized by a few scattered oaks and a grass-weed ground cover. It is best developed on the south slopes where the soil material is deeper than that found in the pine-andropogon community. The oak-grass community is found on the No. 9 and on the Newdale Deposits.

On the No. 9 Deposit *Quercus imbricaria* is the dominant tree. *Q. stellata*, *Q. alba*, and *Q. falcata* are the sub-dominant trees. The oaks are 6 inches to 2 feet in diameter and up to 30 ft. tall. Most of them have gnarled branches, and some of them have many dead branches in the tops. *Ceanothus americanus*, *Vaccinium stamineum*, *Kalmia latifolia*, *Smilax glauca*, and *S. rotundifolia* var. *quadrangularis* are the common shrubs scattered through the community. *Panicum lanuginosum* var. *fasciculatum*, *P. Boscii* var. *molle*, *P. sphaerocarpon*, *P. dichotomum*, *Thalictrum revolutum*, *Asclepias verticillata*, *Anychia polygonoides*, *Lysimachia quadrifolia*, *Lespedeza hirta*, *Danthonia spicata*, *Scleria ciliata*,

Sisyrinchium gramineum, *Euphorbia corollata*, *Verbascum Thapsus*, and *Silene antirrhina* are the common herbaceous species.

The oak-grass community on the south slope of the Newdale Deposit has a large barren rock outcrop with a few weeds in the small joints between the rocks. The trees are confined to the region around the barren area and to the larger joints between the rocks. *Quercus stellata* and *Q. borealis* var. *maxima* are the dominant trees. These oaks are 6–15 inches in diameter and 30–40 ft. tall. On the more northerly exposures a few plants of *Acer rubrum* and *Betula lenta* are growing. *Kalmia latifolia*, *Corylus americana*, *Crataegus macrocarpa*, *Rhododendron calendulaceum*, and *Smilax rotundifolia* var. *quadrangularis* are the common shrubs. The common herbaceous species are *Verbascum Thapsus*, *Veronica officinalis*, *Ambrosia elatior*, *Achillea Millefolium*, *Cirsium lanceolatum*, *Panicum lanuginosum* var. *fasciculatum*, *Digitaria sanguinalis*, *Holcus lanatus*, *Asclepias verticillata*, and *Thalictrum revolutum*.

5) Oak-shrub community

This community is characterized by scattered oaks with dense undergrowth of mostly ericaceous shrubs. It is found on the drier slopes where there is an abundance of soil materials, on the Corundum Hill and Newdale Deposits.

On the Corundum Hill Deposit *Quercus stellata*, *Q. borealis* var. *maxima* and *Oxydendrum arboreum* are the dominant trees. These are 2–6 inches in diameter and 10–20 ft. tall. *Gaylussacia baccata*, *Vaccinium stamineum*, and *Smilax glauca* are the common shrubs. The common herbaceous species are *Panicum lanuginosum* var. *fasciculatum*, *Sisyrinchium mucronatum*, *Hypericum punctatum*, and *Pteridium aquilinum* var. *latiusculum*.

On the Newdale Deposit the dominant trees are *Quercus alba*, *Q. stellata*, and *Q. borealis* var. *maxima*. The oaks are 6–10 inches in diameter and 20–40 ft. tall. *Kalmia latifolia*, *Rhododendron maximum*, *Ceanothus americanus*, *Gaylussacia baccata*, and *Smilax glauca* form a very dense entanglement of shrubs beneath the oaks. In the few open areas between the shrubs are *Panicum lanuginosum* var. *fasciculatum*, *P. sphaerocarpon*, *Hypericum punctatum*, *Hieracium Gronovii*, *Anychia polygonoides*, *Desmodium rhombifolium*, and *Rumex Acetosella*. Most of the olivine boulders in this community are covered with lichens and mosses.

6) Grass-weed community (Pasture on dunite)

This community is composed of native and cultivated grasses, weeds, and a few scattered trees and shrubs. It occurs on Cane Creek, Dark Ridge, Frank, Holcombe Branch, and Newfound Gap Deposits.

The Cane Creek pasture is on a steep slope. There are many olivine boulders in the community with a few grasses and herbs in the crevices between the boulders. *Pinus rigida* and *Crataegus Crus-galli* are scattered throughout the pasture. The pines are 4–6 inches in diameter and 10–15 ft. tall. The dominant herbaceous species are *Agrostis perennans*, *A. hiemalis*, *Paspalum laeve*, *Panicum lanuginosum* var. *fasciculatum*, *Trifolium procumbens*, and *T. repens*. The other

common herbs are *Chrysanthemum Leucanthemum*, *Achillea Millefolium*, *Plantago lanceolata*, *P. virginica*, *Erigeron canadensis*, *Rumex Acetosella*, and *Hedeoma Pulegioides*.

On the Dark Ridge Deposit the grass and herbaceous growth is luxuriant along the streams but poor on the drier areas. Along the railroad tracks through the middle of the pasture, are found *Cornus alternifolia*, *Sassafras albidum*, *Aesculus octandra*, *Hydrangea arborescens*, *Amelanchier canadensis*, *Thuja occidentalis* var. *pyramidalis*, and *Rhododendron calendulaceum*. In the upper end of the pasture is a colony of *Halesia carolina* saplings. *Juglans cinerea*, 12 inches in diameter and 30 feet tall, and *Crataegus Crus-galli* grow in the lower end of the pasture. The herbaceous species are numerous. The ferns *Asplenium platyneuron*, *A. Trichomanes*, *Adiantum pedatum*, *Athyrium asplenoides*, *Osmunda cinnamomea*, *O. regalis* var. *spectabilis*, *O. Claytoniana*, *Polystichum acrostichoides*, and *Dennstaedtia punctilobula* are very common. Some of the other more common herbaceous species are *Epilobium coloratum*, *Achillea Millefolium*, *Aster divaricatus*, *Barbarea vulgaris*, *Anthoxanthum odoratum*, *Cerastium viscosum*, *Chrysanthemum Leucanthemum*, *Cirsium lanceolatum*, *Clematis Viorna*, *Danthonia spicata*, *Daucus Carota*, *Digitaria sanguinalis*, *Diodia teres*, *Eleusine indica*, *Erigeron annuus*, *E. ramosus*, *Sisymbrium officinale*, *Eupatorium urticaefolium*, *Galinsoga ciliata*, *Gaura biennis*, *Gnaphalium obtusifolium*, *G. purpureum*, *Hedeoma pulegioides*, *Juncus tenuis*, *Lobelia siphilitica*, *Lysimachia quadrifolia*, *Oxalis stricta*, *Plantago lanceolata*, *Phleum pratense*, *Prunella vulgaris*, *Ranunculus recurvatus*, *R. repens*, *Rumex Acetosella*, *Salvia lyrata*, *Sanicula canadensis*, *Silene virginica*, *Specularia perfoliata*, *Trifolium repens*, *Poa compressa*, and *Verbascum Thapsus*.

The pasture on the Frank Deposit occurs on very steep slopes. The western slope is the most barren area found on any of the deposits. The broad-leaved vegetation appears dwarfed and stunted. A few saplings of *Acer rubrum*, *Aesculus octandra*, *Betula lenta*, and *Robinia Pseudo-Acacia* are scattered throughout the community. A few specimens of the following shrubs are found in the pasture: *Kalmia latifolia*, *Spiraea alba*, *Rhododendron maximum*, *R. calendulaceum*, *Crataegus collina*, *C. Crus-galli*, *Alnus rugosa*, *Smilax glauca*, and *S. rotundifolia* var. *quadrangularis*. The common herbaceous species are *Adiantum pedatum*, *Aquilegia canadensis*, *Achillea Millefolium*, *Agrostis alba*, *A. hiemalis*, *Anychia polygonoides*, *Arenaria serpyllifolia*, *Asclepias verticillata*, *Silene virginica*, *Dianthus Armeria*, *Eupatorium urticaefolium*, *Gnaphalium obtusifolium*, *Lepidium virginicum*, *Lobelia inflata*, *Sorghastrum nutans*, *Verbascum Blattaria*, *V. Thapsus*, and *Veronica officinalis*.

The pasture on the Holcombe Branch Deposit is the only one cut for hay and is reseeded frequently. A small part of this pasture is fenced and grazed. Very few large boulders are found in the entire pasture, which is located on gentle slopes. A few trees, *Acer rubrum*, *Prunus virginiana*, *Oxydendrum arboreum*, *Cornus florida*, *Juglans nigra*, *Pinus Strobus*, *Crataegus macrosperma*, and *C. Crus-galli* occur in the pasture. The trees are 6-12 inches in diameter, 15-50 ft. tall. The dominant herbs are *Agrostis alba*, *Holcus lanatus*, *Phleum pratense*, *Bromus commutatus*, *Trifolium repens*, *Achillea Millefolium*, *Dactylis glomerata*,

Ambrosia elatior, *Chrysanthemum Leucanthemum*, *Poa compressa*, *Erigeron annuus*, *Lepidium virginicum*, *Plantago lanceolata*, *Rumex Acetosella*, and *Veronica officinalis*.

The upper third of the pasture on the Newfound Gap Deposit is wooded by oaks; the center third is devoid of trees, and the lower third is wooded by black locusts. The common trees are *Quercus borealis* var. *maxima*, *Q. alba*, *Q. montana*, *Q. velutina*, *Q. coccinea*, *Robinia Pseudo-Acacia*, *Carya glabra*, and *Pinus virginiana*. The oaks are 9–15 inches in diameter, and 30–50 ft. tall. The black locusts are 12 inches in diameter and 50–60 ft. tall. Most of the latter are dead or dying. *Ceanothus americanus*, *Hydrangea arborescens*, *Rubus argutus*, and *Rubus allegheniensis* are the common shrubs. A large colony of *Rhus glabra* covers an area about 30 feet wide and 75 feet long beneath the power lines near the top of the deposit. These shrubs are about 8 feet tall. The more common herbaceous species are *Gerardia tenuifolia*, *Agrostis perennans*, *Dactylis glomerata*, *Danthonia spicata*, *Muhlenbergia Schreberi*, *Panicum commutatum*, *P. dichotomum*, *Phleum pratense*, *Pteridium aquilinum* var. *latiusculum*, *Rumex Acetosella*, *Sphenopholis obtusata*, *Carex rosea*, *Sisyrinchium gramineum*, *Rudbeckia hirta*, and *Silene virginica*. *Senecio aureus* forms a large ground cover in the center third of the community. *Nepeta hederacea* makes a carpet beneath the black locusts on the lower third. Lichens are abundant on the trees and rocks in this community.

7) Alder-sedge-smartweed community

This community is found along the banks of streams that cross the deposits. It occurs on the Frank, Holcombe Branch, and Newdale Deposits.

On the Frank Deposit several saplings of *Platanus occidentalis* and *Populus candicans* are scattered along the stream. *Alnus rugosa* is the most common shrub. *Sambucus canadensis*, *Salix sericea*, *S. nigra*, *Rosa palustris*, and *Symphoricarpos orbiculatus* are the other less common shrubs. The common herbaceous species are *Carex lurida*, *C. vulpinoidea*, *Leersia oryzoides*, *Juncus effusus*, *J. marginatus*, *Eleocharis obtusa*, *Polygonum Hydropiper*, *P. pennsylvanicum*, *P. sagittatum*, *Scirpus sylvaticus*, *Solidago canadensis*, *Equisetum arvense*, *Ranunculus acris*, *Lycopus uniflorus*, *Helenium autumnale*, *Oenothera biennis*, *Mentha piperita*, *Mimulus ringens*, *Lysimachia Nummularia*, and *Myosotis laxa*.

On the Holcombe Branch Deposit, several saplings of *Platanus occidentalis* are found along the stream. The common shrubs are *Alnus rugosa*, *Benzoin aestivale*, *Sambucus canadensis*, *S. pubens*, *Salix sericea*, *S. nigra*, *Rosa carolina*, *Rubus allegheniensis*, *R. occidentalis*, *Vitis bicolor*, *V. cordifolia*, and *V. labrusca*. *Carex lurida*, *C. vulpinoidea*, *Glyceria striata*, *Leersia oryzoides*, *Juncus tenuis*, *J. effusus*, *Hypericum mutilum*, *Helenium autumnale*, *Eleocharis obtusa*, *Apios tuberosa*, *Ludvigia palustris*, *Lysimachia Nummularia*, *Mentha piperita*, *Lycopus virginicus*, *Onoclea sensibilis*, *Scirpus atrovirens*, *S. sylvaticus*, *Solidago canadensis*, *Polygonum hydropiperoides*, *P. pennsylvanicum*, *P. virginianum*, *Rudbeckia laciniata*, *Eupatorium purpureum*, *E. perfoliatum*, *Cryptotaenia canadensis*, *Echinochloa crus-galli*, *Cicuta maculata*, *Aster paniculatus*, *Lactuca spicata*, and *L. villosa* are the common herbaceous species on this deposit, in the alder-sedge-smartweed community.

Saplings of *Betula lenta*, *Carpinus caroliniana*, *Liriodendron tulipifera*, and *Prunus virginiana* occur along the stream on the Newdale Deposit. The common shrubs are *Alnus rugosa*, *Rhododendron calendulaceum*, *Kalmia latifolia*, and *Sassafras albidum*. *Carex scoparia*, *C. debilis*, *C. lurida*, *C. vulpinoidea*, *Cyperus strigosus*, *Helenium autumnale*, *Juncus effusus*, *J. tenuis*, *Lobelia cardinalis*, *Lycopus virginicus*, *Muhlenbergia Schreberi*, *Panicum clandestinum*, *Polygonum pennsylvanicum*, *P. Persicaria*, *P. punctatum*, *P. sagittatum*, *Scirpus atrovirens*, *S. validus*, and *Cardamine pennsylvanica*, are the common herbaceous species.

8) *Grass-weed-oak community* (Pasture on soils derived from rocks surrounding the olivine deposits).

This community is developed on soils derived from gneiss, biotite schist, and quartz surrounding the olivine deposits. The vegetation is composed predominantly of grass and herbaceous weeds, but in general more trees and shrubs are found in this community than in the grass-weed community on dunite. The pastures surrounding the Cane Creek and Newfound Gap Deposits were studied.

Pinus rigida, *P. echinata*, *Quercus alba*, *Q. montana*, *Q. falcata*, and *Q. stellata* are the common trees found in the community surrounding Cane Creek. These trees are 4–8 inches in diameter and 15–30 ft. tall. The more common shrubs are *Sambucus canadensis*, *Rhododendron calendulaceum*, *Benzoin aestivale*, *Calycanthus nanus*, *Rhus copallina*, and *Hydrangea arborescens*. *Agrostis perennans*, *Dactylis glomerata*, *Panicum dichotomum*, *Paspalum laeve*, *Hedeoma pulegioides*, *Gnaphalium obtusifolium*, *Cassia nictitans*, *Erigeron annuus*, *E. pulchellus*, *Polystichum acrostichoides*, *Rumex Acetosella*, *Trifolium pratense*, *T. procumbens*, *T. repens*, *T. agrarium*, *Plantago lanceolata*, *Antennaria plantaginifolia*, and *Krigia virginica* are the common herbaceous species.

In the community surrounding the Newfound Gap Deposit, *Quercus alba*, *Q. montana*, *Pinus virginiana*, *Acer rubrum*, and *Carya glabra* are the dominant trees. These are 6–12 inches in diameter and 20–50 ft. tall. The common shrubs are *Castanea pumila*, *Benzoin aestivale*, *Hydrangea arborescens*, *Amorpha fruticosa*, and *Rhododendron calendulaceum*. *Agrostis alba*, *Panicum lanuginosum* var. *fasciculatum*, *Achillea Millefolium*, *Poa compressa*, *Danthonia spicata*, *Trifolium repens*, *Veronica arvensis*, *Prunella vulgaris*, *Nepeta hederacea*, *Holcus lanatus*, *Hypoxis hirsuta*, *Juncus tenuis*, *Plantago aristata*, *Pteridium aquilinum* var. *latiusculum*, *Specularia perfoliata*, *Solanum carolinense*, and *Erigeron pulchellus* are the common herbs.

FLORISTICS

General

The vascular flora on the ten deposits is represented by 750 species and varieties. The maximum number on any single deposit is 344, on the Dark Ridge Deposit. The minimum number is 162, on the Cane Creek Deposit. The maximum number in any one community is 454, in the maple-beech-birch-magnolia community. The minimum, 106, is in the oak-shrub community. A total of 3475 specimens was collected on the ten deposits during this study.

The vascular flora consists of 712 angiosperms (537 dicotyledons and 175 monocotyledons), 8 gymnosperms, and 30 pteridophytes. The trees are represented by 74 species, the shrubs by 53 species, the woody vines by 11, and the herbs by 612. For the number of species on each deposit and in each community, see Table IV.

The following species and varieties are found on all ten deposits: *Asplenium platyneuron*, *Polystichum acrostichoides*, *Smilax rotundifolia* var. *quadrangularis*,

TABLE IV
Species Summary

DEPOSITS	CC	CH	DB	DR	D9	F	HB	LC	N	NG	TOTAL
Vascular Plants.....	162	233	233	344	182	219	249	199	195	177	750
Pteridophytes.....	9	14	12	19	11	13	6	14	6	8	30
Gymnosperms.....	3	1	4	3	3	1	4	3	1	2	8
Angiosperms.....	150	218	217	322	168	205	239	182	188	167	712
Monocotyledons.....	33	58	55	63	32	53	43	40	48	29	175
Dicotyledons.....	117	160	162	259	136	152	196	142	140	138	537
Trees.....	25	23	28	35	25	20	32	24	22	21	74
Shrubs.....	13	30	19	31	16	17	25	25	16	16	53
Woody Vines.....	1	3	6	4	1	1	5	1	2	2	11
Herbs.....	121	177	180	274	140	181	187	149	155	138	612

COMMUNITIES	mbm	og	os	pa	pas	pd	po	s	TOTAL
Vascular Plants.....	454	181	106	171	176	351	248	229	750
Pteridophytes.....	25	9	7	9	9	17	16	7	30
Gymnosperms.....	3	4	1	3	4	6	4	1	8
Angiosperms.....	426	168	98	159	163	334	228	221	715
Monocotyledons.....	108	31	26	39	31	62	51	53	175
Dicotyledons.....	318	137	72	120	132	272	177	168	537
Trees.....	46	26	15	21	27	43	36	20	74
Shrubs.....	38	17	19	27	15	24	25	17	53
Woody Vines.....	7	1	1	1	2	6	5	4	11
Herbs.....	363	137	71	122	132	284	182	188	612

Rubus argutus, *Hypericum punctatum*, *Kalmia latifolia*, *Rhododendron calendulaceum*, and *Erigeron ramosus*. Following are those found on nine of the deposits: *Adiantum pedatum*, *Panicum dichotomum*, *P. lanuginosum* var. *fasciculatum*, *Poa compressa*, *Smilax glauca*, *Phytolacca americana*, *Cerastium viscosum*, *Silene virginica*, *Liriodendron tulipifera*, *Sassafras albidum*, *Amphicarpa monoica*, *Acer rubrum*, *Ceanothus americanus*, *Sanicula canadensis*, *Solanum carolinense*, *Verbascum Thapsus*, *Plantago lanceolata*, *Chrysanthemum Leucanthemum*, and *Erigeron pulchellus*.

The maple-beech-birch-magnolia community occurs on six of the deposits. *Kalmia latifolia* and *Erigeron ramosus* are found in this community on all six.

The following species are found in this community on five of the deposits: *Adiantum pedatum*, *Polystichum acrostichoides*, *Chamaelirium luteum*, *Hydrangea arborescens*, *Acer rubrum*, *Lysimachia quadrifolia*, and *Eupatorium urticaefolium*.

The pine-andropogon community is found on four deposits. Plants found on all four are: *Quercus stellata*, *Lespedeza hirta*, *Ceanothus americanus*, *Hypericum punctatum*, *Chimaphila maculata*, *Kalmia latifolia*, and *Coreopsis major*. The following species are found in the pine-andropogon community on three of the deposits: *Pinus rigida*, *Panicum dichotomum*, *Lilium carolinianum*, *Thalictrum revolutum*, *Sassafras albidum*, *Robinia Pseudo-Acacia*, *Oxydendrum arboreum*, *Chrysopsis mariana*, *Erigeron ramosus*, *Hieracium venosum*, and *Sericocarpus asteroides*.

The pine-oak community occurs on four of the deposits. The following species are found on all four: *Asplenium platyneuron*, *Quercus borealis* var. *maxima*, *Kalmia latifolia*, and *Rhododendron calendulaceum*. The following species are found in this community on three of the deposits: *Polystichum acrostichoides*, *Pteridium aquilinum* var. *latiusculum*, *Pinus Strobus*, *P. virginiana*, *Iris cristata*, *Quercus stellata*, *Thalictrum revolutum*, *Euphorbia corollata*, *Acer rubrum*, *Ceanothus americanus*, *Lyonia ligustrina*, *Oxydendrum arboreum*, *Vaccinium pallidum*, and *Solanum carolinense*.

The oak-grass community is found on two of the deposits. The following plants occur in this community on both of the deposits: *Adiantum pedatum*, *Asplenium platyneuron*, *Polystichum acrostichoides*, *Panicum dichotomum*, *P. sphaerocarpon*, *Smilax rotundifolia* var. *quadrangularis*, *Quercus alba*, *Q. stellata*, *Phytolacca americana*, *Anemone virginiana*, *Thalictrum revolutum*, *Sassafras albidum*, *Platanus occidentalis*, *Rubus argutus*, *Robinia Pseudo-Acacia*, *Acer rubrum*, *Hypericum punctatum*, *Psedera quinquefolia* var. *hirsuta*, *Sanicula canadensis*, *Cornus alternifolia*, *Kalmia latifolia*, *Asclepias verticillata*, *Solanum carolinense*, *Verbascum Thapsus*, *Specularia perfoliata*, *Erigeron ramosus*, *Eupatorium aromaticum*, *Gnaphalium obtusifolium*, and *Solidago nemoralis*.

The oak-shrub community is found on two of the deposits. The following species are found in this community on both deposits: *Fagus grandifolia*, *Quercus stellata*, *Rubus argutus*, *R. occidentalis*, *Ceanothus americanus*, *Hypericum punctatum*, *Sanicula canadensis*, *Nyssa sylvatica*, *Kalmia latifolia*, *Rhododendron calendulaceum*, *Scutellaria pilosa*, *Galium circaezans*, *Houstonia purpurea*, and *Aster divaricatus*.

The grass-weed community is found on five deposits. *Panicum lanuginosum* var. *fasciculatum*, *Poa compressa*, and *Erigeron ramosus* are found on all five deposits. The following species are found in this community on four of the deposits: *Smilax glauca*, *S. rotundifolia* var. *quadrangularis*, *Rumex Acetosella*, *Phytolacca americana*, *Silene virginica*, *Lepidium virginicum*, *Rubus argutus*, *Robinia Pseudo-Acacia*, *Trifolium repens*, *Acer rubrum*, *Hypericum punctatum*, *Cornus florida*, *Kalmia latifolia*, *Rhododendron maximum*, *Verbascum Thapsus*, *Veronica officinalis*, *Plantago lanceolata*, *Specularia perfoliata*, *Achillea Millefolium*, *Gnaphalium purpureum*, and *Senecio Smallii*.

The alder-sedge-smartweed community is found on three of the deposits.

Species found on all three are: *Echinochloa crus-galli*, *Holcus lanatus*, *Carex lurida*, *C. vulpinoidea*, *Cyperus strigosus*, *Scirpus atrovirens*, *Juncus effusus*, *Salix nigra*, *Alnus rugosa*, *Polygonum pennsylvanicum*, *P. sagittatum*, *Rubus argutus*, *Lycopus virginicus*, *Achillea Millefolium*, *Ambrosia elatior*, *Chrysanthemum Leucanthemum*, *Erigeron ramosus*, *Helenium autumnale*, and *Vernonia noveboracensis*.

The grass-weed-oak communities around two deposits were studied. Following are the species found in both: *Asplenium platyneuron*, *Athyrium thelypteroides*, *Polystichum acrostichoides*, *Pteridium aquilinum* var. *latiusculum*, *Juniperus virginiana*, *Panicum lanuginosum* var. *fasciculatum*, *Poa compressa*, *Arisaema atrorubens* forma *viride*, *Juncus tenuis*, *Carya glabra* var. *villosa*, *Quercus alba*, *Q. montana*, *Q. velutina*, *Polygonum Persicaria*, *Rumex Acetosella*, *R. obtusifolius*, *Phytolacca americana*, *Cerastium vulgatum*, *Ranunculus recurvatus*, *Benzoin aestivale*, *Lepidium virginicum*, *Hydrangea arborescens*, *Potentilla canadensis*, *Rubus argutus*, *Trifolium repens*, *Hypericum punctatum*, *Sanicula canadensis*, *Rhododendron calendulaceum*, *Solanum carolinense*, *Verbascum Thapsus*, *Plantago lanceolata*, *Houstonia purpurea*, *Specularia perfoliata*, *Erigeron pulchellus*, *E. ramosus*, *Gnaphalium purpureum*, and *Senecio Smallii*.

Endemism

Mason (1946), in discussing the reasons for endemism in California states: "It is obvious that the Napa-Lake area owes its high concentration of endemics to its great edaphic diversity and especially to the fact that this diversity involves peculiar rocks and their associated minerals. Next in order of importance is the presence in the area of a number of genera capable of elaboration over these peculiar habitats. Those genera which give rise to an occasional endemic species are not responsible for the large number of endemics. It is, on the other hand, such genera as *Navarretia*, *Linum*, and *Streptanthus* which develop many local species and habitat races, that build up the population of restricted endemics in a small area and that give the impression of some sort of genetic instability. It becomes clear that diversity in the environment must work with genetic diversity and must involve diverse tolerances, to produce situations such as that in the Napa-Lake area or in any rich flora." Fernald (1907), in his study of the soil preferences of alpine plants, found 21 species strictly localized on the serpentine of Mt. Albert, Gaspé County, Quebec. I have found none of those species on the North Carolina or Georgia deposits. Pennell (1910) found 17 species restricted to the Conowingo Barrens (serpentine) in southeastern Pennsylvania. Of those 17 species, *Sphenopholis obtusata* and *Asclepias verticillata* are found on the olivine deposits I have studied.

I have not found any species that I consider endemic to the olivine deposits in North Carolina or Georgia on which I have collected extensively. Several species that are localized on the deposits are not found in the immediate areas surrounding the deposits, but do occur elsewhere in the state. These species are as follows: *Quercus stellata*, *Pinus rigida*, *Thalictrum revolutum*, *Asclepias verticillata*, *Physocarpus opulifolius*, *Danthonia spicata*, *Andropogon scoparius*, *A.*

furcatus, *Cystopteris fragilis*, *Camptosorus rhizophyllus*, *Carex plantaginea*, *C. leptalea*, *C. heterosperma*, *Cyperus filiculmis* var. *macilentus*, *Scleria ciliata*, *S. oligantha*, *Aletris farinosa*, *Sisyrinchium gramineum*, *S. mucronatum*, *S. angustifolium*, *Arenaria serpyllifolia*, *A. patula*, *Lespedeza hirta*, *Polygala Senega*, *Liatris graminifolia*, *L. spicata*, *Clematis Beadlei*, *Ruellia humilis*, *Eupatorium latidens*, and *Frasera caroliniensis*.

Range Extensions

Arenaria patula Muhl., *Ruellia humilis* Nutt., and *Eupatorium latidens* Small are reported as new to North Carolina. *Clematis Beadlei* (Small) Erickson is reported as new to Georgia.

New county locations for the following ferns in North Carolina are:

Jackson County—*Polypodium polypodioides*

Avery County—*Camptosorus rhizophyllus*

Osmunda cinnamomea

Yancey County—*Adiantum pedatum*

Asplenium Trichomanes

Athyrium thelypteroides

Dennstaedtia punctilobula

Dryopteris marginalis

Onoclea sensibilis

Polystichum acrostichoides var. *incisum*

Plants of Interest

Plants found on the olivine deposits that normally grow in calcareous regions are *Camptosorus rhizophyllus*, *Ruellia humilis*, *Arenaria patula*, *Frasera caroliniensis*, and *Carex plantaginea*.

Two coastal plain species found on the deposits are *Scleria ciliata* and *S. oligantha*.

Some of the rarer species collected were *Botrychium alabamense*, *Nemexia Hugerii*, *Epipactis repens* var. *ophioides*, *Liparis Loeselii*, *Hydrangea cinerea*, *Ilex Beadlei*, and *Evonymus obovatus*.

EXPLANATION OF CATALOG

In the catalog I have followed as closely as possible the Engler and Prantl System as revised by Robinson and Fernald in "Gray's New Manual of Botany," 1908. The most recent taxonomic publications have been used in determining the species and varieties. The deposits, and the communities on the deposits in which each species or variety was found, follow each species or variety listed in the catalog. Deposits are abbreviated by capital letters, and communities by small letters. For example:

Kalmia latifolia L.

DR: mbm, po

indicates that *Kalmia latifolia* L. was found on the Dark Ridge Deposit in maple-beech-birch-magnolia and pine-oak communities. The abbreviations of the deposits and the communities, and their meanings as employed in the catalog, have been explained on page 53.

A question mark following a plant name in the catalog indicates that the specimen lacks flowers or fruits for definite identification. An asterisk before the name indicates that the species is introduced.

CATALOG OF VASCULAR PLANTS

POLYPODIACEAE

Adiantum pedatum L.

CC: pas; DR: mbm, pd, po; HB: s; CH: og, pa; D9: mbm, og; LC: mbm; DB: mbm; F: mbm, pd; NG: pd.

Asplenium platyneuron (L.) Oakes

CC: pas, pd; D9: mbm, og; N: og; CH: os, pas; F: mbm, pd; NG: pas, pd; DB: mbm, pa, po; HB: pa, pd, po; DR: mbm, po; LC: po.

Asplenium Trichomanes L.

DR: pd, po; F: mbm; N: og; D9: mbm; LC: mbm, po.

Athyrium asplenoides (Michx.) Eaton

CC: pas; DR: mbm, pd; N: s; CH: og; D9: mbm; NG: pas, pd; DB: mbm; LC: mbm.

Athyrium thelypteroides (Michx.) Diels

CH: os; D9: mbm; N: s; DR: po; LC: po.

Camptosorus rhizophyllus (L.) Link

DR: mbm; F: mbm.

Cheilanthes lanosa (Michx.) Eaton

LC: po.

Cystopteris fragilis (L.) Bernh.

DR: mbm, po; F: mbm, pd; HB: s.

Dennstaedtia punctilobula (Michx.) Moore

DB: mbm, po; F: pd; N: og; DR: pd; LC: po.

Dryopteris hexagonoptera (Michx.) C. Chr.

CC: pas; D9: mbm; NG: pd; DR: mbm, pd, po; LC: mbm, po.

Dryopteris marginalis (L.) A. Gray

CH: pa; DR: mbm, pd, po; NG: pd; DB: mbm.

Dryopteris noveboracensis L.

DR: mbm, pd; LC: mbm, pa.

Dryopteris spinulosa (Retz.) Kuntze var. *intermedia* Underwood

CH: pa; F: mbm.

Onoclea sensibilis L.

DB: mbm; HB: s.

Polypodium polypodioides L.

CC: pas; CH: pa; LC: mbm, po.

Polypodium virginianum L.

DB: mbm; D9: og; F: mbm; DR: mbm, pd, po.

Polystichum acrostichoides (Michx.) Schott

CC: pas, pd; D9: mbm, og; N: og, s; CH: os, pa; F: mbm, pd; NG: pas, pd;
DB: mbm, pa, po; HB: pd, po, s; DR: mbm, pd; LC: mbm, pa, po.

Polystichum acrostichoides (Michx.) Schott var. *incisum* Gray

DB: mbm.

Pteridium aquilinum L. var. *latiusculum* (Desv.) Underwood

CC: pas; DR: mbm; LC: mbm, po; CH: os, pa; F: mbm; NG: pas, pd;
DB: mbm, po; HB: po.

Woodsia obtusa (Spreng.) Torr.

CC: pas, pd; DR: pd; D9: og.

Woodwardia areolata (L.) Moore

CH: mbm.

OSMUNDACEAE

Osmunda cinnamomea L.

CH: os; F: pd; NG: pd; DR: mbm, pd.

Osmunda Claytoniana L.

CH: mbm; DR: pd; F: mbm.

Osmunda regalis L. var. *spectabilis* Willd.

CH: os; DR: mbm, pd; LC: mbm, po; DB: mbm; D9: og.

SELAGINELLACEAE

Selaginella apoda L.

CC: pas.

OPHIOGLOSSACEAE

Botrychium alabamense Maxon

LC: pa, po.

Botrychium dissectum Sprengel var. *obliquum* (Muhl.) Clute

D9: mbm.

Botrychium dissectum Sprengel var. *tenuifolium* (Underwood) Farr.

CH: mbm.

Botrychium virginianum (L.) Swartz

CH: mbm, os; DR: mbm, pd; D9: mbm; DB: po.

EQUISETACEAE

Equisetum arvense L.

F: s.

PINACEAE

Juniperus virginiana L.

CC: pas; D9: mbm, og; NG: pas.

Pinus echinata Mill.

CC: pas, pd.

Pinus pungens Lamb.

HB: pd.

Pinus rigida Mill.

CC: pas, pd; DB: pa, po; HB: pa, po; CH: pa; DR: po.

Pinus Strobus L.

DB: mbm, pa, po; LC: mbm, po; N: og, os; HB: pa, pd, po.

Pinus virginiana Mill.

DB: po; HB: po; NG: pas, pd; D9: og; LC: pa, po.

**Thuja orientalis* L. var. *pyramidalis* Endl.

DR: pd.

Tsuga canadensis (L.) Carr.

DB: mbm; D9: og; LC: mbm, po; DR: mbm; F: s.

ALISMACEAE

Sagittaria latifolia Willd. var. *pubescens* (Muhl.) J. G. Sm.

DR: pd; F: s.

GRAMINEAE

**Agropyron repens* (L.) Beauv.

DB: mbm.

**Agrostis alba* L.

DB: mbm, pa; F: pd; NG: pas; HB: pd, s; N: s.

Agrostis hiemalis (Walt.) B.S.P.

CC: pd; CH: mbm; F: pd.

Agrostis perennans (Walt.) Tuckerm.

CC: pas, pd; HB: po, s; NG: pd; DB: po; LC: mbm; DR: mbm, pd; N: os.

Andropogon furcatus Muhl.

CH: pa; DB: po; NG: pd.

Andropogon scoparius Michx.

CH: os; F: pd; N: og; DB: mbm, po; HB: pa, pd.

**Anthoxanthum odoratum* L.

CH: mbm, pa; DR: pd; D9: og.

Aristida longespica Poir.

F: s.

**Arrhenatherum elatius* (L.) Mert. & Koch

N: og.

Arundinaria tecta (Walt.) Muhl.

CH: mbm, pa; LC: mbm.

Brachelytrum erectum (Schreb.) Beauv.

DR: mbm; LC: mbm.

**Bromus commutatus* Schrad.

DB: mbm; HB: pd, s; NG: pd; DR: mbm.

**Bromus secalinus* L.

DR: mbm.

Cinna arundinacea L.

CH: mbm; DB: po.

**Dactylis glomerata* L.

CC: pas; HB: pd; NG: pd; F: s; N: og.

Danthonia sericea Nutt.

CH: pa; LC: po.

Danthonia spicata (L.) Beauv.

CH: pa, os; D9: og; N: s; DR: mbm, pd, po; F: mbm; NG: pas, pd.

**Digitaria ischaemum* (Schreb.) Muhl.

CC: pas; DB: po; F: mbm, s.

**Digitaria sanguinalis* (L.) Scop.

CC: pd; DR: pd; N: og, s.

Echinochloa colona (L.) Link

HB: s.

**Echinochloa crus-galli* (L.) Beauv.

DB: po; F: s; N: og, s; DR: pd; HB: s.

**Eleusine indica* (L.) Gaertn.

CC: pas; HB: pd; N: os; DR: pd.

Eragrostis capillaris (L.) Nees.

CC: pd; HB: s; N: og, os, pd.

Festuca elatior L.

DR: pd; NG: pas.

Festuca obtusa Spreng.

HB: s.

Festuca octoflora Walt.

D9: og.

Glyceria melicaria (Michx.) F. T. Hubb

DR: pd; NG: pd.

Glyceria striata (Lam.) Hitchc.

CH: mbm, pa; DR: mbm, pd; F: s; DB: mbm; D9: mbm; HB: s.

**Holcus lanatus* L.

DB: mbm, pa, po; F: s; NG: pas, pd; DR: pd; HB: po, s; D9: mbm; N: og, s.

Hystrix patula Moench.

DR: mbm.

Leersia oryzoides (L.) Swartz.

HB: s; F: s.

Leersia virginica Willd.

CC: pas; DR: pd; F: s; DB: po.

**Lolium multiflorum* Lam.

LC: po.

Melica mutica Walt.

LC: mbm, pa, po.

Muhlenbergia Schreberi Gmel.

F: pd, s; N: s; NG: pd.

Panicum auburne Ashe

CH: os; DB: mbm; NG: pas.

Panicum Boscii Poir.

CH: os; DR: mbm, pd, po; LC: mbm, po; DB: pa; D9: mbm.

Panicum Boscii Poir. var. *molle* (Vasey) Hitchc. & Chase

DB: po; D9: og; HB: po.

Panicum capillare L.

N: og, s.

Panicum clandestinum L.

DB: mbm; HB: pa, s; N: s; DR: mbm.

Panicum commutatum Schult.

CH: os; LC: mbm, pa, po; NG: pd.

Panicum commutatum Schult. var. *Ashei* (Pearson) Fernald

CH: pa; D9: mbm; N: os; DB: pa; HB: po; DR: mbm; LC: mbm, po.

Panicum depauperatum Muhl.

CH: os; LC: po.

Panicum dichotomum L.

CC: pas; DR: mbm; LC: mbm, po; CH: pa; D9: og; N: og; DB: pa; HB: pa, po; NG: pd.

Panicum lanuginosum Ell. var. *fasciculatum* Fernald

CC: pas, pd; DR: mbm, pd; HB: pd, s; CH: mbm, os; D9: mbm, og; LC: pa, po; DB: pa, po; F: mbm, pd; NG: pas, pd.

Panicum lanuginosum Ell. var. *septentrionale* Fernald

LC: mbm.

Panicum linearifolium Scribn.

CH: mbm, pa; LC: mbm, pa.

Panicum microcarpon Muhl.

CH: mbm; DB: mbm; N: s.

Panicum minus (Muhl.) Nash

DB: po.

Panicum sphaerocarpon Ell.

CH: mbm, pa; HB: pa; N: og; D9: og; LC: po.

Paspalum ciliatifolium Michx.

CC: pas.

Paspalum laeve Michx.

CC: pas.

Paspalum pubescens Muhl.

CC: pd; F: s; HB: s.

**Phleum pratense* L.

DB: mbm; F: pd; NG: pas, pd; DR: mbm, pd; HB: po.

**Poa annua* L.

NG: pd.

**Poa compressa* L.

CC: pas, pd; DR: pd; HB: pd; CH: mbm; D9: mbm; N: og, pd; DB: mbm; F: mbm; NG: pas, pd.

**Poa pratensis* L.

DR: pd.

Poa sylvestris Gray

CH: pa; NG: pas, pd.

**Setaria lutescens* (Weigel) F. T. Hubb

CC: pas; HB: pd; N: og, os.

Sorghastrum nutans (L.) Nash

CC: pd; F: pd.

Sphenopholis nitida (Spreng.) Scribn.

D9: mbm; N: os.

Sphenopholis obtusata (Michx.) Scribn.

DR: mbm; NG: pd.

**Sporobolus Berteroanus* (Trin.) H. & C.

N: s.

Stipa avenacea L.

CH: mbm; LC: mbm, pa, po.

Triodia flava (L.) Smyth

CC: pas; D9: mbm, og; HB: pd; CH: mbm.

Trisetum pennsylvanicum (L.) Beauv.

CH: mbm; LC: po.

Uniola latifolia Michx.

LC: po.

CYPERACEAE

Carex acutivalis M. A. Curtis

F: mbm.

Carex blanda Dewey

CC: pas; F: mbm.

Carex Bushii Mackenzie

D9: og.

Carex cephalophora Muhl.

CC: pas; DB: pa; D9: mbm, og; CH: mbm; DR: pd; N: s.

Carex digitalis Willd.

DB: mbm; F: mbm; LC: po.

Carex flexuosa Muhl.

N: s.

Carex Frankii Kunth.

HB: s.

Carex grisea Wahl.

HB: s.

Carex gynandra Schw.

D9: og.

Carex heterosperma Wald.

N: os.

Carex hirsutella Mackenzie

CH: mbm, os; D9: mbm, og.

Carex incompta Bickn.

DB: mbm; DR: pd.

Carex laxiflora Lam.

D9: og; LC: mbm.

Carex laxiflora Lam. var. *gracillima* Boott

DR: pd; D9: mbm.

Carex leptalea Wahl.

DB: mbm.

Carex lucorum Willd.

F: s.

Carex lurida Wahl.

CC: pd; DR: pd; N: s; CH: mbm; F: s; DB: mbm; HB: s.

Carex Muhlenbergii Schk.

DB: mbm; N: s.

Carex pennsylvanica Lam.

DB: mbm; DR: pd; F: mbm.

Carex plantaginea Lam.

DR: mbm.

Carex rosea Schk.

CH: mbm; DR: mbm, pd; N: s; DB: mbm; D9: mbm; NG: pd.

Carex rosea Schk. var. *radiata* Dewey

NG: pd.

Carex scabrata Schw.

CH: mbm; DR: mbm; LC: po.

Carex scoparia Schk.

CH: os; N: s.

Carex stricta Lam.

F: s.

Carex varia Muhl.

DB: pa; D9: mbm; LC: po; DR: mbm; F: mbm; N: os.

Carex virescens Muhl.

CH: mbm; LC: mbm.

Carex vulpinoidea Michx.

CH: mbm; F: s; N: s; HB: s.

Cyperus cylindricus (Ell.) Britton

CC: pas.

Cyperus echinatus (Ell.) Wood

CC: pd.

Cyperus filiculmis Vahl var. *macilentus* Fernald *

CC: pd.

Cyperus flavescens L.

CC: pas; HB: s; N: s.

Cyperus strigosus L.

CC: pas; DR: pd; N: s; CH: mbm, pa; F: s; DB: pa; HB: s.

Eleocharis acicularis (L.) R. & S.

F: s.

Eleocharis obtusa (Willd.) Schultes

CC: pas; F: s; HB: s; CH: mbm.

Fimbristylis autumnalis (L.) R. & S.

DR: pd; F: s.

Rhynchospora capitellata (Michx.) Vahl.

CH: mbm; F: s.

Scirpus atrovirens Muhl.

DB: pa; HB: s; N: s; F: s

Scirpus Cyperinus (L.) Kunth

DB: pa.

Scirpus lineatus Michx.

CH: mbm.

Scirpus polyphyllus Vahl.

HB: s.

Scirpus sylvaticus L.

F: s; N: s.

Scirpus validus Vahl.

N: s.

Scleria ciliata Michx.

D9: og; HB: pa; N: og.

Scleria oligantha Michx.

LC: mbm, po.

ARACEAE

Arisaema atrorubens (Ait.) Blume forma *viride* (Engler) Fernald

CC: pas; D9: mbm; NG: pas, pd; CH: mbm, os; F: mbm, pd; DR: mbm, pd, po; LC: po.

Arisaema atrorubens (Ait.) Blume forma *zebrinum* (Sims) Fernald

F: mbm.

Arisaema triphyllum (L.) Schott var. *montanum* Fernald

CC: pas.

COMMELINACEAE

Commelina communis L.

DB: mbm; N: s; NG: pd; D9: mbm.

Tradescantia subaspera Ker-Gawl var. *montana* (Shuttlew.) Anderson & Woodson

DB: po; DR: mbm, pd, po; F: pd.

JUNCACEAE

Juncus acuminatus Michx.

CC: pd; HB: s.

Juncus aristulatus Michx.

DB: mbm.

Juncus canadensis J. Gay

F: s; N: s.

Juncus canadensis J. Gay var. *subcaudatus* Engelm.

CH: mbm.

Juncus effusus L.

CH: mbm; F: s; N: s; DB: mbm; HB: s.

Juncus marginatus Rostk.

DB: mbm; DR: po; F: s.

Juncus secundus Beauv.

HB: pa, pd.

Juncus tenuis Willd.

CC: pas; DR: mbm, pd; N: os, s; CH: mbm; D9: og; NG: pas, pd; DB: pa, po; HB: s.

Luzula campestris (L.) DC. var. *bulbosa* A. Wood

CH: mbm, os; DR: pd; NG: pas, pd; DB: po.

Luzula saltuensis Fernald

LC: mbm, po.

LILIACEAE

Aletris farinosa L.

CH: pa; DR: mbm.

Chamaelirium luteum (L.) A. Gray

CH: mbm, pa; F: pd; LC: mbm, po; DR: mbm.

Clintonia umbellulata Michx.

DR: mbm; F: mbm.

Disporum lanuginosum (Michx.) Nichols

DR: mbm.

Erythronium americanum Ker. ?

F: mbm.

**Hemerocallis fulva* L.

HB: s.

Lilium carolinianum Michx.

CH: pa; DR: mbm; HB: pa, po; DB: pa.

Medeola virginiana L.

DB: mbm; LC: mbm.

Nemexia herbacea (L.) Small

DB: mbm; DR: mbm, pd.

Nemexia Hugerii Small

CH: os; D9: mbm, og.

Oakesia puberula (Michx.) Wats.

DR: mbm; LC: mbm; N: os; F: mbm.

Polygonatum biflorum (Walt.) Ell.

DB: mbm; D9: mbm; LC: po; DR: mbm.

Polygonatum commutatum (Schultes) A. Dietrich

DB: mbm, po; DR: mbm.

Smilacina racemosa Desf.

DB: mbm, pa, po; D9: og; F: mbm; DR: mb

Smilax glauca Walt.

CC: pas; DR: mbm, pd; HB: pd; CH: mbm, os, pa; D9: mbm, og; LC: mbm, po; DB: pa, po; F: pd; NG: pd.

Smilax rotundifolia L. var. *quadrangularis* (Muhl.) Wood

CC: pd; D9: og; N: og; CH: mbm, os; F: mbm, pd; NG: pas; DB: mbm, pa; HB: pa, pd; DR: pd, po; LC: mbm.

Stenanthium gramineum (Ker.) Morong

CH: pa; DB: mbm, pa, po.

Trillium Catesbaei Ell.

CH: mbm.

Trillium erectum L.

DR: mbm.

Trillium erectum L. var. *album* (Michx.) Pursh

DB: mbm.

Trillium Gleasoni Fernald

CH: mbm; DR: mbm.

Trillium Gleasoni Fernald x *T. Vaseyi* Harbison var. *simile* Gleason

D9: mbm.

Uvularia grandiflora J. E. Smith

DR: mbm.

Yucca filamentosa L. var. *concava* Baker ?

LC: po.

DIOSCOREACEAE

Dioscorea glauca Muhl.

NG: pd.

Dioscorea villosa L.

DR: mbm, pd, po.

AMARYLLIDACEAE

Hypoxis hirsuta (L.) Coville

CH: pa; LC: mbm; NG: pas; DR: mbm; N: os.

IRIDACEAE

Iris cristata Ait.

CH: mbm; DB: po; LC: po; DB: po; F: mbm.

Sisyrinchium angustifolium Mill.

CH: pa; DR: pd.

Sisyrinchium gramineum Curtis

DB: mbm; N: s; NG: pas, pd; LC: po.

Sisyrinchium mucronatum Michx.

CH: mbm, pa; D9: og; LC: pa, po.

ORCHIDACEAE

Aplectrum hyemale (Muhl.) Torr.

CH: mbm; DR: mbm.

Corallorhiza odontorhiza Nutt.

LC: mbm.

Cypripedium acaule Ait.

HB: po.

Cypripedium parviflorum Salisb.

F: mbm.

Epipactis pubescens (Willd.) A. A. Eaton

F: mbm; LC: mbm; N: os.

Epipactis repens (L.) Crantz var. *ophioides* (Fernald) A. A. Eaton

DB: mbm.

Habenaria ciliaris (L.) R. Br.

CH: mbm; DR: po.

Habenaria clavellata (Michx.) Spreng.

CH: mbm.

Habenaria lacera (Michx.) R. Br.

NG: pd.

Liparis liliifolia (L.) L. C. Rich

CH: os; D9: mbm; LC: mbm; DR: mbm.

Liparis Loeselii (L.) L. C. Rich

DR: mbm.

Orchis spectabilis L.

F: mbm.

Spiranthes gracilis (Bigel.) Beck

CC: pas; D9: mbm; LC: mbm, pa, po; CH: pa; F: pd; N: og.

Spiranthes tuberosa Raf.

HB: pd.

Tipularia discolor (Pursh) Nutt.

LC: mbm.

SALICACEAE

**Populus candicans* Ait.

DB: mbm; F: s.

Salix humilis Marsh

CH: mbm.

Salix nigra Marsh

CC: pas; F: s; N: s; CH: mbm; HB: s.

Salix sericea Marsh

CC: pd; F: s; HB: pd, s; DB: mbm.

JUGLANDACEAE

Carya alba (L.) K. Koch

CC: pas; DR: mbm, po; LC: mbm; DB: mbm.

Carya glabra (Mill) Spach

CC: pd; NG: pas, pd.

Carya glabra (Mill) Spach var. *villosa* (Sarg.) Robinson

CC: pas; D9: mbm; NG: pas; CH: pa.

Juglans cinerea L.

DR: mbm, pd.

Juglans nigra L.

CC: pas, pd; HB: pd, s; LC: mbm.

BETULACEAE

Alnus rugosa (Du Roi) Spreng.

CH: os, pa; F: pd, s; N: s; DR: pd; HB: s.

Betula lenta L.

CH: os; F: mbm, pd; NG: pas; DB: mbm, po; HB: pa; D9: mbm; N: og, os, s.

Betula lutea Michx.

DB: mbm; DR: mbm.

Carpinus caroliniana Walt.

DR: mbm, pd; F: mbm; N: s; D9: mbm, og.

Corylus americana Walt.

CH: os; D9: mbm, og; N: og; DR: mbm.

Corylus rostrata Ait.

HB: po; N: og.

Ostrya virginiana (Mill) Willd.

HB: po.

FAGACEAE

Castanea dentata (Marsh) Borkh.

DB: pa; D9: mbm; NG: pd; DR: mbm, po; HB: po.

Castanea pumila (L.) Mill

DB: pa; HB: pa, po, s; N: os; D9: og; LC: po; NG: pas, pd.

Fagus grandifolia Ehrh.

CH: os; F: mbm; LC: po; DB: mbm; HB: po; N: os.

Quercus alba L.

CC: pas, pd; DR: mbm; N: og; CH: os; D9: mbm, og; NG: pas, pd; DB: mbm, pa, po; LC: mbm.

Quercus borealis Michx. f. var. *maxima* (Marsh) Ashe

CC: pd; F: mbm; N: og, s; DB: pa, po; HB: po; NG: pas, pd; DR: mbm, po; LC: po.

Quercus coccinea Muench.

CH: pa, os; D9: mbm, os; NG: pd; DB: mbm, pa; F: pd; DR: mbm; N: po.

Quercus falcata Michx.

CC: pas; D9: mbm, og; LC: pa, po; CH: pa; HB: pa, pd, po.

Quercus imbricaria Michx.

D9: mbm, og.

Quercus montana Willd.

CC: pas; DB: mbm; D9: mbm; CH: os; DR: mbm; NG: pas, pd.

Quercus stellata Wang.

CC: pd; DR: mbm; LC: pa, po; CH: os, pa; D9: og; N: og, os; DB: mbm, pa, po; HB: pa, pd, po, s.

Quercus velutina Lam.

CC: pas; DB: po; NG: pas, pd; CH: os; N: og, os.

URTICACEAE

Boehmeria cylindrica (L.) Sw.

DB: po.

Laportea canadensis (L.) Gaud.

CC: pas; DB: mbm; DR: mbm.

Machura pomifera (Raf.) Schneider

HB: s.

Morus rubra L.

HB: pd, po; DR: mbm; D9: mbm.

Pilea pumila (L.) Gray

HB: s; F: s.

Ulmus fulva Muhl.

DR: mbm.

SANTALACEAE

Pyrularia pubera Michx.

CH: mbm; D9: og; N: os; DB: mbm; HB: po; DR: mbm; LC: po.

ARISTOLOCHIACEAE

Aristolochia macrophylla Lam.

DR: mbm, pd; F: mbm.

Asarum canadense L.

DR: mbm.

Asarum grandiflorum (Michx.) Small

LC: mbm, po.

POLYGONACEAE

Polygonum buxiforme Small

DR: pd.

Polygonum dumetorum L.

CC: pas; F: s; NG: pd; DR: pd; HB: s.

Polygonum erectum L.

HB: pd.

Polygonum Hydropiper L.

CC: pas; F: s; NG: pd; HB: pd; N: og.

Polygonum hydropiperoides Michx.

DR: pd; HB: s.

Polygonum neglectum Besser

N: s.

Polygonum pennsylvanicum L.

DR: pd; HB: s; N: s; F: s.

**Polygonum Persicaria* L.

CC: pas; F: mbm; N: og, s; DB: po; HB: pd, s; NG: pas.

Polygonum punctatum (Ell.) Small

LC: mbm; N: s.

Polygonum sagittatum L.

DB: po; F: s; N: s; DR: pd; HB: s.

Polygonum tenue Michx.

DR: pd.

Polygonum virginianum L.

CC: pas; DR: pd; NG: pd; CH: mbm; HB: s.

**Rumex Acetosella* L.

CC: pas, pd; DR: pd; N: og, os; CH: mbm; F: pd, s; NG: pas, pd; DB: mbm, po; HB: pd, po, s.

**Rumex crispus* L.

CH: mbm; D9: mbm; N: s.

**Rumex obtusifolius* L.

CC: pas; DR: mbm, pd; NG: pas, pd; DB: mbm; HB: pd, s.

CHENOPODIACEAE

**Chenopodium ambrosioides* L.

CC: pas; F: pd; N: og; DR: pd; HB: po.

**Chenopodium album* L.

DR: po; N: og, s; NG: po; HB: pa, pd.

AMARANTHACEAE

**Amaranthus hybridus* L.

DR: pd; N: og, os, s.

PHYTOLACCACEAE

Phytolacca americana L.

CC: pas; DR: pd; HB: pa, pd, po; CH: os; D9: og; N: og, s; DB: mbm, pa, po; F: pd, s; NG: pas, pd.

ILLECEBRACEAE

Anychia canadensis (L.) B. S. P.

DB: mbm; NG: pd.

Anychia polygonoides Raf.

DB: pa, po; F: pd, s; N: og; D9: og.

CARYOPHYLLACEAE

Arenaria patula Michx.

DR: pd.

Arenaria serpyllifolia L.

F: mbm, pd, s; N: og; NG: pd.

**Cerastium viscosum* L.

CC: pd; D9: og; N: os; CH: mbm; F: mbm, s; NG: pd; DR: mbm, pd;
HB: po.

**Cerastium vulgatum* L.

CC: pas, pd; N: og, os; NG: pas, pd; F: s.

**Dianthus Armeria* L.

CH: mbm; F: pd, s; NG: pas, pd; DB: mbm, pa, po; HB: pd.

**Lychnis alba* Mill

F: s.

Sagina decumbens (Ell.) T. & G. var. *Smithii* (Gray) Wats.

LC: po.

**Saponaria officinalis* L.

HB: s.

Silene antirrhina L.

DR: pd; D9: og.

Silene noctiflora L.

NG: pd.

Silene stellata (L.) Ait. f.

F: mbm.

Silene virginica L.

CC: pas, pd; D9: mbm, og; LC: po; DB: po; F: pd, s; NG: pd; DR: mbm,
pd; HB: pa.

Stellaria graminea L.

DB: mbm.

Stellaria media (L.) Cyrill.

DR: pd; NG: pd.

Stellaria pubera Michx.

DR: mbm, pd.

PORTULACACEAE

Portulaca oleracea L.

DB: mbm.

RANUNCULACEAE

Aconitum uncinatum L.

D9: mbm.

Actaea alba (L.) Mill

DR: mbm; LC: mbm, po.

Anemone virginiana L.

DB: mbm; HB: po; NG: pd; DR: mbm, pd; LC: mbm; D9: mbm, og; N: og.

Aquilegia canadensis L.

F: mbm.

Cimicifuga racemosa (L.) Nutt.

CH: mbm; D9: mbm; HB: po; DR: mbm, pd.

Clematis Beadlei (Small) Erickson

LC: mbm.

Clematis Viorna L.

DR: pd; F: pd.

Clematis virginiana L.

DB: mbm, po; F: s; NG: pd.

**Delphinium Ajacis* L.

F: s.

Hepatica acutiloba DC.

DR: mbm; F: mbm.

Ranunculus abortivus L.

NG: pd.

**Ranunculus acris* L.

F: s; N: po, s.

Ranunculus hispidus Michx.

NG: pd.

Ranunculus recurvatus Poir.

CC: pas, pd; DR: mbm, pd; HB: s; CH: mbm; D9: mbm; NG: pas, pd;

DB: mbm; F: s.

Ranunculus repens L.

DR: pd.

Ranunculus sceleratus L.

N: os.

Thalictrum clavatum DC.

DB: mbm; DR: mbm; F: s.

Thalictrum dioicum L.

CH: mbm.

Thalictrum polygamum Muhl.

DB: po.

Thalictrum revolutum DC.

CH: mbm, pa, os; DR: mbm, po; HB: pa, po; DB: mbm, pa, po; D9: mbm,

og; N: og.

Zanthorhiza apiifolia L'Her.

CH: mbm, pa; LC: mbm, po; N: s.

MAGNOLIACEAE

Liriodendron tulipifera L.

CC: pas; DR: mbm, pd, po; LC: po; CH: os; D9: mbm, og; N: s; DB: pa;

HB: pd; NG: pd.

Magnolia acuminata L.

DB: mbm, pa, po; DR: mbm, pd, po; D9: mbm, og.

Magnolia Fraseri Walt.

DR: mbm; F: mbm; LC: mbm, po.

CALYCANTHACEAE

Calycanthus fertilis Walt.

CH: pa; DR: mbm.

Calycanthus nanus (Loisel) Small

CC: pas; DR: mbm.

ANONACEAE

Asimina triloba (L.) Dunal

LC: mbm.

BERBERIDACEAE

Caulophyllum thalictroides L.

CC: pas; DR: mbm, pd.

Podophyllum peltatum L.

CC: pd; D9: mbm; NG: pd; CH: os; LC: mbm; DR: mbm; N: os.

LAURACEAE

Benzoin aestivale (L.) Nees.

CC: pas; DR: po; N: pas CH: os; D9: og; NG: pd; DB: mbm; HB: pd, s.

Sassafras albidum (Nutt.) Nees.

CC: pas; DR: mbm, pd; LC: mbm, po; CH: pa, os; D9: mbm, og; N: og, s;

DB: mbm, pa, po; HB: pa, po, s; NG: pd.

PAPAVERACEAE

Sanguinaria canadensis L.

CC: pas; D9: mbm; NG: pd; CH: mbm, pa; F: mbm, pd; DR: mbm, pd;

LC: po.

CRUCIFERAE

**Arabidopsis thaliana* (L.) Britton

HB: s.

Arabis canadensis L.

CH: os; DR: mbm; NG: pas, pd.

Arabis laevigata (Muhl.) Poir.

DR: pd.

**Barbarea verna* (Mill) Aschers

HB: s.

**Barbarea vulgaris* R. Br.

DR: pd.

**Brassica japonica* Siebold

DR: pd.

**Brassica nigra* (L.) Koch.

DR: pd.

**Brassica rapa* L.

N: s.

**Capsella Bursa-pastoris* (L.) Medic

CC: pas; DR: pd; N: os.

Cardamine pennsylvanica Muhl.

N: s.

Dentaria heterophylla Nutt.

DR: mbm.

**Lepidium campestre* (L.) R. Br.

HB: pa; N: og, s.

Lepidium virginicum L.

CC: pas, pd; DR: mbm, pd; N: og, os, s; CH: mbm; F: pd, s; NG: pas;

DB: pa; HB: pd, po.

**Sisymbrium altissimum* L.

DR: pd.

**Sisymbrium officinale* (L.) Scop.

CC: pas; HB: pd; NG: pas; DR: pd; N: og, s.

**Thlaspi arvense* L.

DR: pd.

CRASSULACEAE

Sedum ternatum Michx.

DB: mbm; HB: s; NG: pas, pd; F: mbm.

SAXIFRAGACEAE

Astilbe biternata (Vent.) Britton

CH: mbm; DR: mbm; D9: mbm; DB: mbm.

Heuchera americana L.

CH: mbm; HB: a; N: og.

Heuchera villosa Michx.

DR: mbm; F: pd; LC: mbm.

Hydrangea arborescens L.

CC: pas; D9: mbm; NG: pas, pd; CH: mbm, pa; F: mbm; DR: mbm, pd.

LC: mbm.

Hydrangea cinerea Small

LC: mbm.

Mitella diphylla L.

CC: pas; DR: mbm.

Tiarella cordifolia L.

DR: mbm; LC: mbm, po.

HAMAMELIDACEAE

Hamamelis virginiana L.

DB: mbm; F: mbm, pd; LC: mbm; DR: mbm, po.

Liquidambar styraciflua L.

LC: mbm, po.

PLATANACEAE

Platanus occidentalis L.

CC: pd; D9: og; HB: pd, s; CH: og; F: s.

ROSACEAE

Agrimonia parviflora Soland.

CC: pas; DB: po; HB: pd, po, s; CH: pa.

Agrimonia rostellata Wallr.

NG: pd.

Agrimonia striata Michx.

DR: pd; D9: og.

Amelanchier canadensis (L.) Medic.

HB: pa, po; LC: po.

Amelanchier laevis Wiegand

CH: mbm; DR: pd.

Aruncus allegheniensis Rydb.

DB: mbm; F: s.

Aruncus Aruncus (L.) Karst.

CH: mbm.

**Chaenomeles lagenaria* (Loisel) Koidz.

F: s.

Crataegus collina Chapm.

HB: s; F: pd.

Crataegus Crus-galli Chapm.

CC: pd; F: s; HB: pd, s; DR: pd.

Crataegus macrosperma Ashe

F: pd; LC: po; NG: pas; HB: pd, s; N: og, s.

Fragaria virginiana Duchesne

CH: mbm; DR: pd; NG: pas, pd.

Geum canadense Jacq.

CC: pas; DR: mbm; NG: pd, s; DB: mbm; HB: pd, s.

Geum hirsutum Muhl.

DR: pd.

Gillenia trifoliata Moench.

CH: pa; DR: mbm, pd; LC: mbm; DB: po.

Malus angustifolia (Ait.) Michx.

CH: os; LC: po; N: og.

Malus coronaria (L.) Mill.

CC: pd; F: pd.

Malus glabrata Rehder

HB: po; NG: pd.

**Malus Malus* (L.) Britton

DB: mbm, pa; HB: pa; NG: pas, pd; D9: og.

Malus platycarpa Rehder

DB: pa; HB: pd.

Physocarpus opulifolius (L.) Maxim.

CH: pa, os.

Potentilla canadensis L.

CH: pas, pd; NG: pas, pd.

Potentilla monspeliensis L.

DR: mbm, pd; HB: s; NG: pd.

Potentilla pumila Poir.

CC: pd; DR: pd.

Potentilla recta L.

DR: pd.

Prunus americana Marsh

D9: og; HB: s.

**Prunus Avium* L.

HB: s.

**Prunus Cerasus* L.

DR: pd.

**Prunus Persica* (L.) Stokes

CH: os.

Prunus virginiana L.

CH: mbm; F: mbm; NG: pd; DB: mbm; HB: pa, pd, s; D9: og; N: s.

Rosa carolina L.

CH: pa; HB: s.

**Rosa damascena* Mill

DR: pd.

Rosa palustris Marsh

D9: mbm; F: s.

Rosa setigera Michx. var. *tomentosa* T. & G.

HB: po.

Rubus allegheniensis Porter

CH: pa; DR: mbm; NG: pas, pd; DB: pa; F: pd, s.

Rubus argutus Link

CC: pas, pd; D9: mbm, og; N: og, os, s; CH: mbm, pa, os; F: s; NG: pas, pd; DB: mbm, pa; HB: pd, s; DR: pd; LC: po.

Rubus canadensis L.

DB: pa.

Rubus flagellaris Willd.

CH: pa, os; N: og; NG: pas; LC: po.

Rubus occidentalis L.

CH: os; D9: mbm, og; N: os; DB: mbm, pa, po; HB: po, s; NG: pd; DR: mbm, pd; LC: mbm.

Rubus odoratus L.

DR: mbm; F: pd.

Spiraea alba Du Roi

F: pd.

Waldsteinia fragarioides (Michx.) Tratt.

LC: mbm, po.

LEGUMINOSAE

Amorpha fruticosa L.

NG: pas, pd.

Amphicarpa monoica (L.) Ell.

CC: pas; DR: mbm; HB: s; CH: mbm; D9: mbm; LC: mbm; DB: po; F: s; N: s.

Apios tuberosa Moench.

DB: po; F: mbm; HB: s.

Baptisia tinctoria (L.) R. Br.

CH: mbm, pa.

Cassia chamaecrista Chapm.

CH: mbm.

Cassia nictitans L.

CC: pas; LC: mbm; N: og; DB: po.

Centrosema virginianum (L.) Benth.

LC: po.

Desmodium canadense (L.) DC.

DB: po.

Desmodium ciliare DC.

CH: mbm; HB: pa; LC: po; D9: mbm.

Desmodium laevigatum (Nutt.) DC.

DB: po; HB: po.

Desmodium marilandicum (L.) DC.

DB: po.

Desmodium nudiflorum (L.) DC.

DR: mbm; D9: mbm; LC: mbm, pa.

Desmodium paniculatum (L.) DC.

CC: pd; DR: pd; NG: pd.

Desmodium paniculatum (L.) DC. var *pubens* T. & G

CH: mbm; HB: pd; NG: pd; DB: po.

Desmodium rhombifolium (Ell.) DC.

DB: po; N: og.

Desmodium rigidum (Ell.) DC.

CH: pa; DR: po; LC: mbm; DB: pa.

Desmodium rotundifolium (Michx.) DC.

LC: pa.

Desmodium viridiflorum (L.) Beck.

CH: mbm; LC: mbm.

Gleditsia triacanthos L.

HB: po.

Lespedeza hirta (L.) Ell.

CH: pa; DR: mbm; HB: pa; DB: pa, po; D9: og; LC: pa.

Lespedeza Nuttallii Darl.

DR: mbm; LC: mbm.

Lespedeza procumbens Michx.

CC: pd; D9: mbm, og.

Lespedeza repens (L.) Bart.

LC: pa, pd; D9: og.

Lespedeza virginica (L.) Britton

CH: mbm; HB: s.

**Medicago sativa* L.

HB: pd.

**Melilotus alba* Desv.

HB: pd.

Robinia Pseudo-Acacia L.

CC: pd; DR: mbm; HB: pa, pd, po, s; CH: pa; D9: mbm, og; N: og; DB: mbm, pa, po; F: pd; NG: pas, pd.

Schrankia angustata T. & G.

LC: po.

Stylosanthes biflora (L.) B. S. P.

CH: pa.

Stylosanthes riparia Kearney

CC: pas; D9: mbm, og; N: og; CH: os; HB: po.

**Trifolium agrarium* L.

N: og.

**Trifolium arvense* L.

HB: s.

**Trifolium hybridum* L.

N: s.

**Trifolium pratense* L.

CC: pas; DR: pd; HB: pd, s; DB: mbm, pa; F: s; N: og.

**Trifolium procumbens* L.

CC: pas, pd; D9: og; F: s; CH: mbm.

Trifolium repens L.

CC: pas, pd; D9: mbm; N: s; CH: mbm; F: mbm; NG: pas, pd; DR: pd; HB: pd.

Vicia caroliniana Walt.

CH: pa; DR: pd; F: mbm; DB: po; D9: og; LC: po.

**Vicia sativa* L.

HB: s

LINACEAE

Linum virginianum L.

CH: pa; D9: og; LC: po; DR: mbm; HB: po.

OXALIDACEAE

Oxalis corniculata L.

CH: mbm, pa; DB: mbm.

Oxalis cynnosa Small

CH: pa; F: pd, s; NG: pd; DR: po; N: og.

Oxalis filipes Small

CH: os; N: os.

Oxalis stricta L.

CC: pas, pd; DR: pd; HB: pa; CH: mbm.

Oxalis violacea L.

CC: pas, pd.

GERANIACEAE

Geranium carolinianum L.

CC: pas; CH: mbm; HB: pd, s.

Geranium maculatum L.

D: mbm, pd; F: mbm; NG: pd; D9: mbm.

POLYGALACEAE

Polygala polygama Walt.

LC: mbm, pa, po.

Polygala Senega L.

DR: mbm, pd.

Polygala verticillata L.

CC: pd; D9: og; N: og; DB: pa.

EUPHORBIACEAE

Acalypha rhomboidea Raf.

DR: pd.

Acalypha virginica L.

CC: pd.

Euphorbia corollata L.

CH: pa, os; DR: mbm; HB: pd, po; DB: pa, po; D9: mbm, og; LC: po.

Euphorbia hyssopifolia L.

HB: s; NG: pd.

ANACARDIACEAE

Rhus copallina L.

CC: pas; DR: mbm; LC: po.

Rhus glabra L.

CH: pa; DR: mbm; HB: pd; DB: mbm, po; D9: og; N: s.

Rhus hirta (L.) Sudw.

DB: po.

Rhus Toxicodendron L. var. *radicans* (L.) Torr.

CH: mbm.

AQUIFOLIACEAE

Ilex Beadlei Ashe

CC: pd.

Ilex monticola A. Gray

CH: mbm.

Ilex opaca Ait.

LC: mbm, po.

CELASTRACEAE

Evonymus americanus L.

CH: mbm; D9: mbm; LC: mbm, po; DR: mbm.

Evonymus obovatus Nutt.

DR: mbm.

ACERACEAE

Acer pennsylvanicum L.

DR: mbm; F: mbm.

Acer rubrum L.

CH: pa; D9: mbm, og; LC: mbm, pa, po; DB: mbm, po; F: mbm, pd; N: og, os; DR: mbm, pd; HB: pa, pd, po; NG: pas, pd.

Acer saccharum Marsh

DR: mbm.

SAPINDACEAE

Aesculus octandra L.

CC: pas, pd; F: mbm, pd, s; N: s; DR: pd, po; LC: mbm.

BALSAMINACEAE

Impatiens biflora Walt.

CH: mbm; DB: mbm; HB: s.

Impatiens pallida Walt.

DB: mbm; DR: mbm.

RHAMNACEAE

Ceanothus americanus L.

CH: pa, os; D9: mbm, og; N: os; DB: mbm, pa, po; HB: pa, po; NG: pd; DR: mbm; LC: mbm, pa, po.

MALVACEAE

**Abutilon Abutilon* (L.) Rusby

DR: pd.

**Malva rotundifolia* L.

HB: pd.

HYPERICACEAE

Ascyrum hypericoides L.

CC: pas; DR: pd; LC: pa.

Hypericum canadense L.

DR: pd.

Hypericum mutilum L.

CC: pas; HB: s.

Hypericum perforatum L.

DB: mbm; DR: mbm; HB: s.

Hypericum punctatum Lam.

CC: pas; D9: og; N: og, os, s; CH: pas, os; F: mbm, pd; NG: pas, pd; DB: pa, po; HB: pa, pd, po; DR: mbm, pd; LC: pa.

VIOLACEAE

Viola affinis LeConte

DB: mbm, po; F: mbm; N: og.

Viola blanda Willd.

DR: mbm, pd; LC: po; N: os; F: mbm, s.

Viola canadensis L.

DR: mbm.

Viola cucullata Ait.

CH: mbm; DR: mbm; N: s; DB: po; F: mbm.

Viola eriocarpa Schwein.

DR: mbm.

Viola fimbriatula J. E. Smith

N: og.

Viola hastata Michx.

F: mbm; LC: mbm.

Viola pallens (Banks) Brainerd

DB: mbm.

Viola palmata L.

CH: mbm; LC: po; NG: pd; DR: pd.

Viola papilionacea Pursh

CH: mbm; D9: mbm, og; HB: s.

Viola pedata L.

CH: pa; DR: mbm; HB: po; DB: pa; D9: og; N: og

Viola primulifolia L.

LC: pa, po.

Viola pubescens Ait.

F: mbm.

Viola rotundifolia Michx.

F: mbm; LC: po.

Viola sagittata Ait.

LC: mbm, po.

Viola sororia Willd.

DB: mbm; F: pd.

Viola Walteri House

LC: mbm, po.

PASSIFLORACEAE

Passiflora lutea L.

CC: pas; DR: mbm, pd.

LYTHRACEAE

Cuphea petiolata (L.) Koehne

CH: os; DR: pd; NG: pd.

VITACEAE

Psedera quinquefolia (L.) Greene

CH: mbm; DB: po.

Psedera quinquefolia (L.) Greene var. *hirsuta* (Donn) Rehder

DB: mbm; HB: s; NG: pas; DR: pd; LC: mbm, po; D9: mbm, og; N: og, pd.

Vitis aestivalis Michx.

DB: po; HB: pd.

Vitis bicolor LeConte

DB: mbm; N: os; NG: pas, pd; HB: s.

Vitis cinerea Engelm.

DB: mbm; D9: mbm.

Vitis cordifolia Michx.

CC: pas; DR: pd; HB: pd, po, s; CH: mbm.

Vitis labrusca L.

DB: pa; DR: pd; HB: s.

Vitis rotundifolia Michx.

LC: mbm, po.

TILIACEAE

Tilia neglecta Spach

DR: mbm; F: mbm.

MELASTOMACEAE

Rhexia virginica L.

DR: pd.

ONAGRACEAE

Circaea latifolia Hill

DR: mbm, pd; D9: mbm; NG: pd.

Epilobium coloratum Muhl.

DR: pd; HB: s; NG: pd; F: s

Gaura biennis (L.) Ell.

DB: po; DR: pd; HB: s.

Ludvigia palustris (L.) Ell.

HB: s.

Oenothera biennis L.

CH: mbm; DR: pd; HB: pd, s; DB: mbm; F: s.

Oenothera fruticosa L.

CH: pa.

Oenothera fruticosa L. var. *hirsuta* Nutt.

NG: pas, pd.

Oenothera pumila L.

F: s.

ARALIACEAE

Aralia nudicaulis L.

DR: mbm.

Aralia racemosa L.

DR: mbm.

UMBELLIFERAE

Angelica villosa (Walt.) B. S. P.

DR: mbm; HB: pd; LC: mbm; D9: og.

Cicuta maculata L.

DB: mbm; HB: pd, s.

Cryptotaenia canadensis (L.) DC.

CC: pas; DR: mbm; HB: s; DB: mbm; D9: mbm.

**Daucus Carota* L.

DR: pd; N: og; NG: pas; HB: pd, s.

Eryngium aquaticum L.

CH: pa.

Ligusticum canadense (L.) Britton

LC: mbm.

Osmorhiza Claytoni (Michx.) Clarke

DR: mbm.

Oxypolis rigidior (L.) Coult. & Rose

CH: mbm; DR: mbm, pd; N: s; DB: po.

Sanicula canadensis L.

CC: pas; DR: mbm, pd; LC: mbm; CH: os; D9: mbm, og; N: og, os; DB: mbm; HB: pd, po, s; NG: pas, pd.

Sanicula marilandica L.

DR: mbm.

Sium cicutaeifolium Schrank.

DR: mbm.

Thaspium aureum Nutt.

CH: mbm, os, pa; LC: mbm, po; NG: pas; D9: og.

Thaspium aureum Nutt. var. *atropurpureum* (Desr.) Coult. & Rose

CH: pa; DR: mbm, pd.

Thaspium barbinode (Michx.) Nutt.

DR: mbm.

Zizia Bebbii (Coult. & Rose) Britton

DR: mbm; LC: mbm; NG: pd; F: mbm.

Zizia cordata (Walt.) DC.

DR: mbm; D9: mbm.

CORNACEAE

Cornus alternifolia L.f.

CC: pas; DR: mbm, pd; N: os; DB: mbm, po; LC: po.

Cornus florida L.

CC: pd; D9: mbm, og; N: og; DB: mbm; HB: pd, po, s; NG: pas, pd; DR: mbm, pd; LC: mbm, po.

Nyssa sylvatica Marsh

CC: pas, pd; DR: mbm; N: os; CH: pa, os; D9: og; DB: pa, po; LC: mbm, po.

ERICACEAE

Chimaphila maculata (L.) Pursh

CH: pa, os; HB: pa, po; NG: pas, pd; DB: pa; LC: mbm, pa, po; D9: mbm; N: os.

Clethra acuminata Michx.

CH: pa; DR: mbm; F: mbm.

Epigaea repens L.

DB: pa, po; LC: pa, po; NG: pd; DR: po; N: og, os.

Gaultheria procumbens L.

CH: pa; F: mbm; N: og, os; DR: po.

Gaylussacia baccata Wang.

CH: pa; F: mbm; N: og, os; DR: po; HB: po.

Gaylussacia ursina M. A. Curtis

LC: mbm, po.

Hypopitys americana (DC.) Small

F: mbm; LC: pa.

Hypopitys lanuginosa (Michx.) Nutt.

D9: mbm; LC: po.

Kalmia latifolia L.

CC: pd; D9: mbm, og; N: og, os, s; CH: mbm, pa, os; F: mbm, pd; NG: pas, pd; DB: mbm, pa, po; HB: pa, po; DR: mbm, pd, po; LC: mbm, pa, po.

Leucothoe Catesbaei (Walt.) A. Gray

CH: os; DR: mbm, pd; LC: mbm.

Leucothoe recurva (Buckley) A. Gray

DR: mbm; F: mbm.

Lyonia ligustrina (L.) DC.

CH: pa, os; DR: mbm; LC: pa, po; DB: po; HB: po.

Lyonia ligustrina (L.) DC. var. *foliosiflora* (Michx.) Fernald

DB: mbm; HB: pa; NG: pd.

Monotropa uniflora L.

DB: mbm; DR: mbm.

Oxydendrum arboreum (L.) DC.

CC: pas; DR: mbm; LC: pa, po; CH: pa; D9: mbm, og; N: os; DB: mbm, pa, po; HB: pa, pd, po.

Rhododendron calendulaceum (Michx.) Torr.

CC: pas; D9: og; N: os, s; CH: mbm, os, pa; F: mbm, s; NG: pas, pd; DB: pa, po; HB: po; DR: mbm, pd, po; LC: pa, po.

Rhododendron maximum L.

CH: pa; F: mbm, pd; NG: pd; DB: mbm, pa, po; LC: mbm, po; DR: pd, po; N: og, os.

Vaccinium corymbosum L.

CH: pa.

Vaccinium melanocarpum Mohr.

CH: mbm, pa.

Vaccinium neglectum (Small) Fernald

DB: pa, po; HB: pa; LC: mbm.

Vaccinium pallidum Ait.

CH: mbm; HB: po; LC: mbm, pa, po; DR: po.

Vaccinium stamineum L.

CH: mbm, pa; D9: mbm, og; HB: pa, pd, po; DR: po.

DIAPENSIACEAE

Galax aphylla L.

CH: pa; DR: mbm; LC: mbm, po; DB: mbm, po; F: mbm.

PRIMULACEAE

* *Lysimachia Nummularia* L.

DB: mbm; F: s; HB: s.

Lysimachia quadrifolia L.

CH: mbm, os, pa; DR: mbm, pd; LC: mbm; DB: mbm; D9: mbm, og.

Steironema ciliatum (L.) Raf.

HB: s.

Steironema hybridum (Michx.) Raf.

D9: mbm, og.

EBENACEAE

Diospyros virginiana L.

CC: pas.

STYRACACEAE

Halesia carolina L.

DR: mbm, pd, po.

OLEACEAE

Fraxinus americana L.

DB: mbm; DR: mbm.

Fraxinus pennsylvanica Marsh

D9: mbm.

GENTIANACEAE

Frasera caroliniensis Walt.

D9: og.

Gentiana flavida Gray

D9: og.

Gentiana quinquefolia L.

DR: mbm.

Sabatia angularis (L.) Pursh

CH: pd; D9: mbm, og; N: pd; DR: pd; LC: mbm.

Obolaria virginica L.

D9: mbm; LC: mbm.

ASCLEPIADACEAE

Asclepias exaltata (L.) Muhl.

CH: os.

Asclepias pulchra Ehrh.

F: s.

Asclepias quadrifolia Jacq.

DB: po; DR: mbm.

Asclepias syriaca L.

F: mbm, pd, s.

Asclepias tuberosa L.

DB: mbm; HB: pd; N: og.

Asclepias verticillata L.

DB: po; F: pd; N: og; D9: og; LC: pa.

CONVOLVULACEAE

Cuscuta Gronovii Willd.

DR: mbm, pd; HB: s; NG: pd.

* *Ipomoea purpurea* (L.) Roth.

DR: pd; HB: pd, s.

Ipomoea lacunosa L.

HB: s.

POLEMONIACEAE

Phlox carolina L.

DB: po; D9: mbm; NG: pas, pd; DR: mbm; LC: mbm, pa.

Phlox ovata L.

DB: po.

Phlox stolonifera Sims

CH: mbm; DB: mbm.

HYDROPHYLLACEAE

Hydrophyllum canadense L.

DR: mbm.

BORAGINACEAE

* *Cynoglossum officinale* L.

NG: pas, pd.

Cynoglossum virginicum L.

CH: os; DR: pd; D9: mbm; DB: po.

Myosotis laxa Lehm.

F: s.

VERBENACEAE

Verbena urticaefolia L.

D9: mbm; F: pd; HB: pd.

LABIATAE

Collinsonia canadensis L.

DR: mbm; F: mbm; LC: mbm.

Hedeoma pulegioides (L.) Pers.

CC: pas, pd; DR: pd; N: og; CH: pa; LC: mbm; NG: pd.

Lycopus uniflorus Michx.

F: s.

Lycopus virginicus L.

CC: pas, pd; F: s; N: s; DB: po; HB: s; DR: pd; LC: mbm

* *Mentha piperita* L.

DR: pd; F: s; HB: s.

Monarda clinopodia L.

NG: pd.

Monarda didyma L.

DB: po.

Monarda fistulosa L.

CH: mbm; DR: pd; D9: mbm.

Monarda mollis L.

D9: mbm.

* *Nepeta Cataria* L.

DR: pd; F: pd, s; NG: pd.

* *Nepeta hederacea* (L.) Trevisan

CH: os; F: s; NG: pas, pd; DR: pd.

* *Perilla frutescens* (L.) Britton

LC: mbm.

* *Prunella vulgaris* L.

CH: mbm, os, pa; D9: og; HB: pa, s; DR: pd; F: mbm, pd, s; NG: pas, pd.

Pycnanthemum incanum (L.) Michx.

D9: mbm.

Pycnanthemum montanum Michx.

DR: mbm.

Pycnanthemum pycnanthemoides (Leavenw.) Fernald

CC: pas; F: pd; NG: pd; DR: pd; HB: s; D9: mbm; LC: mbm, po.

Salvia lyrata L.

CH: mbm; DR: pd; D9: mbm, og.

Scutellaria incana Muhl.

CC: pas; D9: mbm.

Scutellaria lateriflora L.

F: s.

Scutellaria pilosa Michx.

CH: os; D9: mbm, og; N: og, os; DB: pa.

Trichostema dichotomum L.

D9: mbm.

SOLANACEAE

**Datura Stramonium* L.

CC: pd; DB: mbm; DR: pd.

**Lycopersicon esculentum* Mill.

N: os.

**Nicandra Physalodes* (L.) Pers.

DB: mbm.

Physalis ambigua (A. Gray) Britton ?

LC: pa.

Physalis angulata L. ?

HB: pd.

Physalis intermedia Rydb. ?

CH: mbm; LC: mbm.

Physalis pubescens L. ?

DR: pd.

Solanum carolinense L.

CC: pas; DR: pd; HB: pa; CH: mbm, pa; D9: og; N: og; DB: pa; F: pd, s;

NG: pas, pd.

Solanum nigrum L.

NG: pd.

SCROPHULARIACEAE

Castilleja coccinea (L.) Spreng.

NG: pas, pd.

Gerardia laevigata Raf.

DR: mbm; LC: mbm, po; N: os; HB: po.

Gerardia purpurea L.

F: s.

Gerardia tenuifolia Vahl.

DR: mbm, pd; D9: mbm; NG: pd.

Melampyrum lineare Desr.

F: mbm; N: os.

Mimulus ringens L.

F: s; HB: s.

Penstemon canescens Britton

DR: pd.

Scrophularia marilandica L.

CC: pas.

**Verbascum Blattaria* L.

CC: pd; F: pd.

**Verbascum Thapsus* L.

CC: pas, pd; D9: og; LC: po; CH: mbm; F: pd, s; N: og; DR: pd; HB: pd;
NG: pas, pd.

Veronica arvensis L.

DR: pd; LC: po; NG: pas, pd; HB: s; N: og.

Veronica officinalis L.

DB: mbm, po; F: mbm, pd; N: og, os; DR: pd; HB: pd, po; NG: pas, pd.

Veronica peregrina L.

DR: pd.

Veronica serpyllifolia L.

CH: mbm; NG: pas.

OROBANCHACEAE

Conopholis americana (L.f.) Wallr.

DB: mbm, po; DR: mbm; LC: mbm.

Epifagus virginiana (L.) Bart.

D9: mbm; F: mbm.

ACANTHACEAE

Ruellia humilis Nutt.

CC: pd.

PLANTAGINACEAE

Plantago aristata Michx.

CH: mbm; F: pd; NG: pas.

**Plantago lanceolata* L.

CC: pas, pd; DR: pd; N: og; CH: mbm; F: s; NG: pas, pd; DB: mbm; HB:
pa, pd.

Plantago Rugelii Dcne.

CC: pas; DR: pd; HB: pd; DB: mbm; F: s; N: s.

Plantago virginica L.

CC: pas, pd.

RUBIACEAE

Diodia teres Walt.

CC: pas, pd; DR: pd; LC: mbm; CH: mbm.

Galium asprellum Michx.

F: s.

Galium circaeans Michx.

CC: pas; DR: mbm, pd; LC: mbm; CH: os; D9: mbm, og; N: os; DB: mbm;
HB: pd.

Galium latifolium Michx.

NG: pd.

Galium pilosum Ait.

D9: mbm, og; NG: pd.

Galium triflorum Michx.

DB: mbm; HB: pd, po, s; NG: pd; F: mbm; LC: mbm.

Houstonia caerulea L.

DB: mbm, pa, po; D9: og; F: mbm, pd, s.

Houstonia purpurea L.

CC: pas, pd; D9: mbm, og; N: os; CH: mbm, pa, os; HB: pa, po; NG: pas,
pd; DR: mbm, pd; LC: mbm, pa, po.

Houstonia purpurea L. var. *pubescens* Britton

D9: og.

Houstonia serpyllifolia Michx.

DR: mbm, pd.

Mitchella repens L.

DB: mbm; LC: mbm, pa, po.

CAPRIFOLIACEAE

**Lonicera japonica* Thunb.

CC: pas.

Sambucus canadensis L.

CC: pas; DR: pd; LC: mbm, CH: mbm; F: pd, s; NG: pd; DB: mbm; HB: s.

Sambucus pubens Michx.

DB: mbm; HB: s; NG: pd.

Symphoricarpos orbiculatus Moench.

D9: mbm; F: s.

Viburnum acerifolium L.

CH: os; DR: mbm; F: mbm.

Viburnum prunifolium L.

HB: pa, po.

CUCURBITACEAE

Sicyos angulatus L.

F: mbm.

CAMPANULACEAE

Campanula americana L.

DR: mbm.

Campanula flexuosa Michx.

CH: mbm, pa, os; F: mbm, pd; NG: pd; DB: mbm; LC: mbm, pa, po; DR: pd; N: og.

Specularia perfoliata (L.) A. DC.

CC: pas, pd; D9: og; N: og; CH: pa; F: pd, s; NG: pas, pd; DR: pd; HB: pa, po, s.

LOBELIACEAE

Lobelia cardinalis L.

DR: mbm, pd; HB: s; N: s.

Lobelia inflata L.

CC: pas; HB: pd; N: og; DB: mbm; LC: mbm.

Lobelia puberula Michx.

CH: pa; D9: og; LC: mbm; DR: mbm.

Lobelia siphilitica L.

DB: mbm; D9: mbm; HB: pd, s; DR: pd; F: s.

Lobelia spicata Lam.

F: mbm, pd.

Lobelia spicata Lam. var. *campanulata* McVaugh

NG: pd.

Lobelia spicata Lam. var. *hirtella* Gray

DB: pa.

Lobelia spicata Lam. var. *leptostachya* (A. DC.) Mackenzie & Bush

D9: og.

Lobelia spicata Lam. var. *originalis* McVaugh

D9: og.

COMPOSITAE

Achillea Millefolium L.

CC: pd; DR: pd; N: og, s; CH: mbm; F: pd, s; Ng: pas, pd; DB: mbm; HB: pa, pd, po, s.

Actinomeris alternifolia DC.

CC: pas; DB: po; D9: mbm.

Ambrosia elatior L.

CC: pas; DR: po; HB: pa, pd, s; CH: mbm, pa; D9: mbm; N: og, s; DB: mbm, po; F: pd, s.

Ambrosia trifida L.

CC: pas; DR: pd; HB: s; DB: po; F: s.

Antennaria plantaginifolia (L.) Richards

CC: pas; NG: pd.

Antennaria solitaria Rydb.

LC: po.

Arctium minus L.

DR: pd; F: s; NG: pd.

Aster Curtisii T. & G.

F: mbm.

Aster divaricatus L.

CH: os; F: mbm, pd, s; N: os; DR: mbm, pd; LC: mbm; NG: pd.

Aster dumosus L.

D9: mbm; LC: pa.

Aster oblongifolius Nutt.

NG: pd.

Aster paniculatus L.

HB: s; N: og.

Aster patens Ait.

DB: mbm, pa, po; HB: pa; N: og.

Aster prenanthoides Muhl.

F: s.

Aster puniceus L.

HB: s.

Aster sagittifolius Willd.

DR: mbm.

Aster undulatus L.

DB: po; DR: mbm; HB: pd.

Bidens bipinnata L.

CC: pas; F: pd; N: og; DR: pd; HB: pd.

Bidens comosa (A. Gray) Wiegand

F: s.

Bidens frondosa L.

HB: s.

Brachyachaeta sphacelata (Raf.) Britton

CC: pas.

Cacalia atriplicifolia L.

DR: mbm; F: pd, s; NG: pas, pd

**Chrysanthemum Leucanthemum* L.

CC: pas, pd; DR: mbm, pd; HB: pd, po, s; CH: mbm; D9: mbm; N: og, os, s; DB: mbm, pa; F: pd, s.

Chrysopsis graminifolia (Michx.) Nutt.

LC: pa.

Chrysopsis mariana (L.) Nutt.

CH: mbm, pa; D9: og; LC: mbm, pa, po; DR: pd; HB: pa, po; NG: pd.

Cichorium Intybus L.

HB: po, s.

**Cirsium lanceolatum* (L.) Hill

DB: po; F: pd; N: og; DR: pd; HB: pd; NG: pd.

Chrysogonum virginianum L.

LC: po.

Coreopsis major Walt.

CH: pa; D9: og; NG: pas; DB: pa, po; HB: pa, po; DR: mbm; LC: pa, po.

**Crepis tectorum* L.

F: s.

Elephantopus carolinianus Willd.

CC: pas.

Elephantopus tomentosus L.

LC: po.

Erechtites hieracifolia (L.) Raf.

DB: mbm, po; DR: pd; HB: pd.

Erigeron annuus (L.) Pers.

CC: pas; D9: mbm; HB: po; DR: pd; F: s.

Erigeron canadensis L.

CC: pas, pd; F: pd; N: og, s; CH: po; HB: po; DR: pd; LC: mbm.

Erigeron pulchellus Michx.

CC: pas; DR: pd; LC: mbm, po; CH: mbm, pa; D9: mbm; N: og; DB: po;
F: pd; NG: pas, pd.

Erigeron ramosus (Walt.) B. S. P. *

CC: pas, pd; D9: mbm, og; N: og, s; CH: mbm, pa, os; F: mbm, pd, s; NG:
pas, pd; DB: mbm, pa, po; HB: pa, pd, po, s; DR: mbm, pd; LC: mbm.

Eupatorium album L.

CH: mbm; D9: mbm.

Eupatorium aromaticum L.

CC: pas; LC: pa; NG: pd; D9: og; N: og, os.

Eupatorium latidens Small

N: pd.

Eupatorium leucolepis T. & G.

CH: os.

Eupatorium maculatum L.

CC: pas; DB: po; HB: s; CH: mbm; D9: mbm; N: s.

Eupatorium perfoliatum L.

DB: po; F: pd, s; HB: s; DR: pd.

Eupatorium sessilifolium L.

CH: os.

Eupatorium trifoliatum L.

DR: mbm; D9: og; LC: mbm.

Eupatorium urticaefolium Reichard

CH: mbm; DR: mbm, pd; F: mbm, pd, s; DB: po; D9: mbm, og; LC: mbm.

**Galinsoga ciliata* (Raf.) Blake

DB: mbm; D9: mbm; N: og, s; DR: pd.

Gnaphalium obtusifolium L.

CC: pas; DR: pd; HB: pd, po; CH: mbm, pa; D9: mbm, og; N: og; DB:
mbm, pa; F: pd.

Gnaphalium purpureum L.

CC: pas, pd; DR: pd; HB: pd; CH: mbm; F: pd; NG: pas.

Helenium autumnale L.

CH: mbm; F: pd, s; N: s; DR: pd; HB: s.

Helenium nudiflorum Nutt.

HB: pd.

Helianthus atrorubens L.

CH: mbm; HB: po; LC: mbm, pa, po; DR: mbm.

Helianthus glaucus Small

CH: pa; D9: mbm, og; LC: pa, po; DR: mbm.

Helianthus tuberosus L.

HB: s.

Hieracium Greenii Porter & Britton

DR: pd; N: os; NG: pas.

Hieracium Gronovii L.

D9: mbm; HB: pd; N: og.

Hieracium paniculatum L.

HB: po.

Hieracium scabrum Michx.

DB: mbm; HB: pa; N: og.

Hieracium venosum L.

CH: pa, os; DR: mbm; LC: pa, po; DB: pa; F: pd; NG: pas, pd.

Krigia virginica (L.) Willd.

CC: pas, pd; DR: pd; N: og.

Lactuca canadensis L.

HB: pd.

Lactuca sagittifolia Ell.

DB: po; HB: po, s.

Lactuca saligna L.

DB: mbm; F: pd; HB: pd.

**Lactuca scariola* L. var. *integrata* Gren. & Godr.

HB: pd; N: og.

Lactuca spicata (Lam.) Hitchc.

DB: po; N: og; NG: pd; HB: pd, s.

Lactuca spicata (Lam.) Hitchc. var. *integrifolia* (Gray) Britton

DR: mbm; HB: s.

Lactuca villosa Jacq.

HB: s.

Liatris graminifolia (Walt.) Willd.

LC: pa.

Liatris graminifolia (Walt.) Willd. var. *Smallii* Gaiser

LC: po.

Liatris spicata (L.) Willd.

CH: pa; DR: mbm, po; LC: pa.

Prenanthes altissima L.

DB: po; DR: mbm; D9: mbm.

Pyrrhopappus carolinianus (Walt.) DC.

HB: s.

Rudbeckia hirta L.

DR: mbm; F: mbm, pd; LC: mbm; D9: mbm; HB: pd; NG: pas, pd.

Rudbeckia laciniata L.

CC: pas; DR: pd; HB: s; DB: mbm, po; F: s.

Senecio aureus L.

DB: mbm; F: s; NG: pd; DR: pd; HB: s.

Senecio Smallii Britton

CC: pas, pd; D9: og; NG: pas, pd; CH: mbm, pa, os; F: pd; DR: pd; LC: po.

Sericocarpus asteroides (L.) B. S. P.

CH: pa, os. HB: pa, po; LC: mbm, pa, po; DR: mbm.

Silphium Asteriscus L.

D9: og.

Solidago bicolor L.

NG: pd.

Solidago Boottii Hook.

CH: pa; HB: po; NG: pd; DR: mbm; LC: mbm, po; F: s; N: os.

Solidago canadensis L.

CH: mbm; D9: og; HB: pd, s; DB: po; F: pd, s; N: s.

Solidago Curtisii T. & G.

DR: mbm; F: mbm.

Solidago erecta Pursh

D9: og; HB: pa, po, s; LC: mbm.

Solidago flexicaulis L.

DR: mbm.

Solidago nemoralis Ait.

CH: pa, po; D9: mbm, og; NG: pd; DB: po; HB: pa, pd; DR: pd; N: og, os.

Solidago odora Ait.

CC: pas; LC: pa, po.

Solidago rigida L.

F: s.

Solidago serotina Ait.

CC: pas; D9: og; NG: pd; DR: pd.

Solidago ulmifolia Muhl.

DB: po.

**Taraxacum officinale* Weber

CC: pas; DR: pd; N: og; CH: mbm; F: pd; NG: pas.

Verbesina occidentalis (L.f.) Walt.

NG: pd.

Vernonia glauca (L.) Britton

F: mbm, pd; HB: pd; N: og.

Vernonia noveboracensis (L.) Willd.

CC: pas; F: s; N: s; CH: mbm; HB: s; NG: pd; DR: pd; LC: mbm.

Xanthium canadensis Mill. ?

HB: s; N: s.

SUMMARY

Ten high-grade olivine deposits in North Carolina and Georgia were studied. Twelve others in the two states were visited during August 1947.

The deposits are characterized by a great number of species but by very few individuals of any one species. 750 species and varieties, which are a representative cross-section of the typical mountain flora of North Carolina, were found on the ten deposits. 3475 specimens were collected and studied. Most of these are deposited in the herbarium of the University of North Carolina.

The communities described are maple-beech-birch-magnolia, pine-andropogon, pine-oak, oak-grass, oak-shrub, grass-weed, alder-sedge-smartweed, and grass-weed-oak. Soil moisture relations are the primary controlling factors in community succession and distribution. Moisture and soil acidity or alkalinity are the controlling factors in species distribution on the deposits.

No correlation between species and mineral distribution on the deposits was discovered. No endemics were found on the deposits. Several species were found localized on the deposits. These were not observed in the areas immediately surrounding the deposits but do occur elsewhere in the state.

Ruellia humilis Nutt., *Eupatorium latidens* Small, and *Arenaria patula* Michx. are reported as new to North Carolina. *Clematis Beadlei* (Small) Erickson is reported as new to Georgia.

Plants found on the olivine deposits that normally grow in calcareous regions are *Campptosorus rhizophyllus*, *Ruellia humilis*, *Arenaria patula*, *Frasera carolinensis*, and *Carex plantaginea*.

ACKNOWLEDGMENTS

This work was done under the direction of Dr. H. R. Totten to whom I wish to express my sincere appreciation for his encouragement and guidance and especially for his invaluable aid in determining species. I also wish to thank Dr. J. M. Fogg of the University of Pennsylvania, and Dr. H. L. Mason of the University of California, for references on serpentine; Dr. J. F. Lutz of State College, Raleigh, N. C., for information on serpentine soils; Dr. W. A. White of the Geology Department of the University of North Carolina for information on rocks, minerals, and soils; Dr. E. C. Markham of the Chemistry Department of the University of North Carolina for determining the pH of soil samples; Mr. Maeburn Huneycutt for aid in collecting; my wife, Laurie Stewart Radford, for assistance in the preparation of the manuscript and plates; and Mrs. Alma Holland Beers for aid in preparing the manuscript for publication.

I also wish to acknowledge grants from the Smith Research Fund, and the Highlands Scholarships, which helped greatly to defray collecting expenses. I am also grateful to the Highlands Museum and Biological Laboratory Inc. for working space and plant drying facilities.

LITERATURE CITED

- FERNALD, M. L. 1907. The Soil Preferences of Certain Alpine and Subalpine Plants. *Rhodora* 9: 149-194.
GORDON, A., AND LIPMAN, C. B. 1930. Why Are Serpentine and Other Magnesian Soils Infertile? *Soil Sci.* 22: 291-302.
HARSHBERGER, JOHN R. 1903. An Ecological Study of Serpentine Barrens of Pennsylvania. *Science* 18: 309-313.

- HUNTER, C. E. 1941. Forsterite Olivine Deposits of North Carolina and Georgia. N. C. Dept. of Conser. and Devel. Bull. No. 41, Raleigh, N. C.
- HURLBUT, C. S. 1941. Dana's Manual of Mineralogy, 15th Ed. John Wiley and Sons Inc., New York.
- MASON, H. L. 1946. The Edaphic Factor in Narrow Endemism. I. The Nature of Environmental Influences. *Madrono* 8: 209-226.
- 1946. The Edaphic Factor in Narrow Endemism. II. The Geographic Occurrence of Plants of Highly Restricted Patterns of Distribution. *Madrono* 8: 241-258.
- MERRILL, G. P. 1921. A Treatise on Rocks, Rock-weathering, and Soils. The Macmillan Co., New York.
- PENNELL, FRANCIS. 1910. Flora of the Conowingo Barrens of Southeastern Pennsylvania. *Proc. Acad. Nat. Sci. of Phila.* 62: 541-585.
- 1912. Further Notes on the Flora of the Conowingo or Serpentine Barrens of Southeastern Pennsylvania. *Proc. Acad. Nat. Sci. of Phila.* 64: 520-542.
- PRATT, J. H., AND LEWIS, J. V. 1905. Corundum and the Peridotites of Western North Carolina. N. C. Geol. Survey Vol. I. Raleigh, N. C.
- ROBINSON, B. L., AND FERNALD, M. L. 1908. Gray's New Manual of Botany. American Book Co., New York, N. Y.
- SCHIMPER, A. F. W. 1903. Plant Geography upon a Physiological Basis. Chapter V. The Soil. Clarendon Press, Oxford, England.

EXPLANATION OF PLATE 8

Top Photograph: CANE CREEK DEPOSIT. Boulders Cover Slopes of Deposits.

Bottom Photograph: BURTON LAKE DEPOSIT. An Example of Oak-Grass Community.
One of the Deposits in Georgia visited during August 1947.

PLATE 8



THE GENUS *Codium* IN NORTH CAROLINA

BY LOUIS G. WILLIAMS

*Department of Botany, Duke University
Durham, North Carolina*

PLATE 9 AND THREE TEXT FIGURES

Among the larger and more conspicuous of the marine algae inhabiting the shallow coastal waters at Beaufort, North Carolina, the genus *Codium* Stackhouse is well represented during the warmer seasons of the year. It is a green alga belonging to the order Siphonales, and according to Smith (1938) is represented in the warmer seas of the world by forty-five species. Collins (1909) lists nine species for North America, Taylor (1928) reports seven species for Florida and Dry Tortugas and Collins (ibid.) and Hoyt (1920), two species for North Carolina.

The macroscopic appearance of *Codium* in North Carolina resembles a small green rope which is commonly dichotomously branched at somewhat regular intervals. The plants vary from perfectly cylindrical to more or less flattened in the portions where branching occurs. Structurally, it is composed of numerous branching, coenocytic filaments, the inner of which are small in diameter, colorless and interwoven into a central core or "medulla." From these inner filaments arise green club-shaped branches called "utricles" which become compactly arranged in a surface layer or "cortex." The utricles function as photosynthetic organs and bear the reproductive organs.

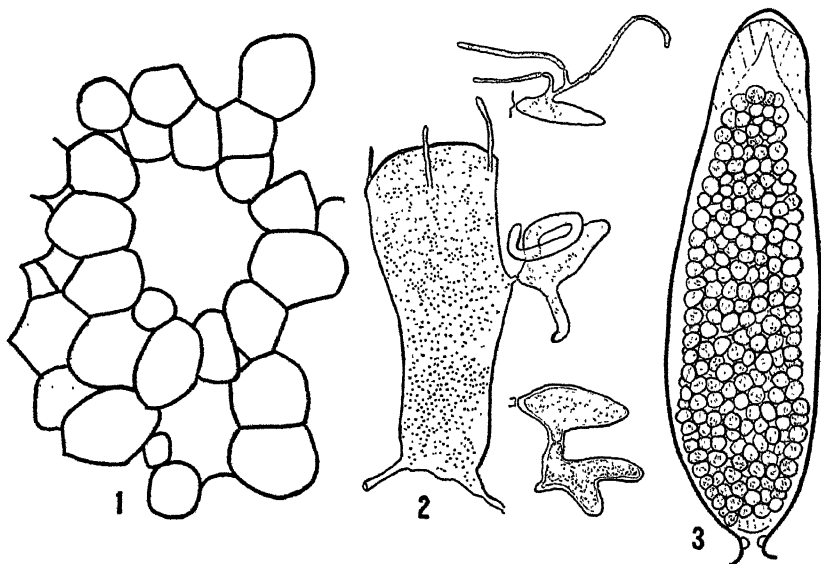
Since little was known about the abundance, distribution, reproduction, and ecology of *Codium* as it occurs in North Carolina, and as there has been some difficulty in distinguishing the two species reported to occur here, an investigation of this alga was commenced in the summer of 1940 and has been continued intermittently up to and through the summer of 1947. Observations have been made on material from Ocracoke to Fort Fisher, but most of the work has been done in the vicinity of Beaufort and Cape Lookout.

The two species of *Codium* reported for Beaufort, North Carolina, are *C. dichotomum* (Huds.) S. F. Gray [reported as *C. tomentosum* (Huds.) Stackh.] and *C. decorticatedum* (Woodward) Howe. The specific differences between these two are mainly in the size of the utricles and the habit of the plants (Plate 9, fig. 2). *Codium dichotomum* has smaller utricles ranging from 84 to 200 μ (usually 130 to 170 μ) in diameter (Collins, 1905; Hoyt, 1920; Schmidt, 1923; and Taylor, 1928) while in *C. decorticatedum* they range from 300 to 400 μ (Hoyt, 1920; Schmidt, 1923; and Taylor, 1928). In habit, *C. dichotomum* is shorter, with terete branches, and little if at all flattened below the dichotomies; *C. decorticatedum* is long and flattened, usually conspicuously so, below the branches.

In discussing the material of *Codium* which occurs at Beaufort, Hoyt (ibid., p. 433) stated that while the forms of both species occur here, he had encountered

considerable difficulty in distinguishing the two species in the Beaufort plants because of the wide variation in each form and the many intergradations between them. Other investigators seem to have experienced similar difficulties with these two species in other localities (Howe, 1911).

In attempting to identify the above species reported to occur here, the author experienced the same difficulty that Hoyt did, that there was too much intergrading of forms to make possible the separation of the two species on habit alone. Therefore, careful and extensive measurements of utricles were undertaken. This revealed that the utricles also have a wide size variation (Text Fig. 1) but that the average for each plant is much larger than in *C. dichotomum*.



Codium decorticateum. Fig. 1. Surface view of portion of a thallus showing giant utricles dispersed among smaller utricles. $\times 25$. Fig. 2. A utricle with one attached propagule and two detached propagules. $\times 55$. Fig. 3. Macrogametangium with macrogametes enclosed. $\times 225$.

It was not unusual to find utricles as large as 575μ in diameter. Furthermore, no correlation of size of utricles could be established between the different forms that occur in the summer and fall. Plants with typical *C. dichotomum* habit had the same size utricles as those of *C. decorticateum*. However, early spring forms of juvenile plants not only had the habit of *C. dichotomum*, but, in some of them, the utricles had the size range for this species, but when growth of these juvenile forms was carefully followed their utricles soon attained the average size of those of *C. decorticateum*. The specimens on which Hoyt based his report were also examined. In comparing the size of utricles in the specimens which he assigned respectively to the two species, no appreciable differences were found in the size of utricles, and all of them fall within the range of those of *C. decorticateum*.

From the above observations it was concluded that upon the basis of size of utricles, which is one of the fundamental characters used in separating the different species of *Codium*, all forms which occur in the Beaufort estuary must be assigned to *C. decorticatedum*.

To check the above conclusion, further study was made upon the habit of the plants. A large number of specimens from different locations were examined. The result of these observations shows that a complete series of intergrading forms exists between the two extreme forms. Further observations also indicate that the *C. dichotomum* form is only a juvenile or depauperate form of *C. decorticatedum*. It is interesting to note that Agardh, in describing *C. elongatum* in 1822 (now *C. decorticatedum*), stated that the juvenile form resembles *C. tomentosum* (now *C. dichotomum*). The most conclusive evidence, however, that the two forms belong to the same species, is the fact that both forms may occur on the same thallus.

Additional evidence that the two forms found here are genetically the same was obtained when tips of both forms were grown under identical, favorable, environmental conditions. It was assumed that they would develop into the same kind of plants if they belonged to the same species, or they would remain unlike if they represented different species. Ninety tips (Plate 9, fig. 3) of each of these two forms respectively were collected and tied to bricks (Plate 9, fig. 4). An effort was made to collect plants from a variety of habitats. After thirty-two days of growth, both sets of tips had tripled in size and had become the long flattened form of *C. decorticatedum* (Plate 9, fig. 5). This experiment furnished further evidence that all of the forms are those of *C. decorticatedum*, and that the form of the plants vary with age and with environmental conditions. Plants growing under favorable environmental conditions become long, flattened, and regularly dichotomously branched. Juvenile, early spring plants and plants growing under poor light conditions develop into short, much-branched thalli, which frequently do not become flattened.

On the basis of the above evidence, it is definitely concluded that all of the plants of *Codium* growing in the Beaufort estuary must be assigned to *C. decorticatedum* and that *C. dichotomum* does not occur here. Below is a description of *C. decorticatedum* as it is represented at Beaufort, North Carolina:

Plants cylindrical, varying from short, more or less irregularly dichotomously branched in stunted and young plants to long and regularly dichotomously branched in large plants which are distinctly flattened, the flattening becoming widest below the branches, where it sometimes reaches nine centimeters; attached by a Vaucheria-like system of colorless rhizoids to firm objects with rough surfaces to a mean depth of twenty-five inches below mean low tide level. Utricles obovate-clavate, with a truncate apex, not thickened or mucronate at apex, 300 to 570 μ in diameter and four to six times as long as the greatest diameter. Plants strictly dioecious. Sporangia (or gametangia) lanceolate-ovate, 230 to 270 μ long by 60 to 100 μ at the greatest diameter, when mature with gelatin inside their tips and bases to a mean depth of 40 μ at the tip, and 25 μ at the base; sporangia of two kinds, equal in size: microsporangia with mature microgametes dark yellow, the gametes small (3-4 x 3 μ after discharge); macrosporangia with

mature macrogametes dark green, the gametes large ($17-18 \times 11-12\mu$ after discharge).

Another species, *C. isthmocladum* Vickers, has occasionally been found drifted ashore in the vicinity of Beaufort. The fresh condition of these plants indicates that they could not have come very far since plants of *Codium* soon degenerate when they become detached and drift. It was not surprising, therefore, to find it attached on the offshore jetty at Cape Lookout. Collections of it at this locality were made in the fall of 1946 and again in the summer of 1947. Here it grew associated with *C. decorticatum*. These two species are not always easily distinguished macroscopically, especially when growing together as young plants and before *C. decorticatum* has developed its characteristic flattening. Microscopic examination of the utricles, however, reveals that the material of *Codium* at Cape Lookout consists of two species. This conclusion is based on the difference in size of utricles and the thickening of the wall of the apex of the utricles. The specimens of *C. isthmocladum* from Cape Lookout have relatively small utricles ($250-300\mu$) in which the outer wall is distinctly thickened at the tip. Also the branches are often constricted at their bases which is not true of *C. decorticatum*. The absence of *C. isthmocladum* in the Beaufort estuary and its presence at Cape Lookout, only 12 miles apart, indicate that environmental conditions of the inshore waters of Beaufort are not suitable for its attachment and growth.

REPRODUCTION

Thuret (1850) seems to have been the first one to see the "gametangia" and biflagellate gametes in *Codium*. Berthold (1880) observed that reproduction did not occur when the male and female plants were grown separately. In September 1896, Oltmanns (1904) saw fusion of anisogametes, which took place between twelve and one o'clock at night. He was able to catch zygotes on glass slides which were placed at the bottom of a *Codium* culture and to observe their germination. In 1925 Miss M. M. Williams—working on *C. dichotomum* in Sydney, Australia—first described meiosis in *Codium*. She found that this took place in the formation of gametes and, therefore, concluded that the plants must be diploid. Previously it had been generally believed that *Codium* was haploid, i.e. a gametophyte, because it produces gametes. This discovery made necessary a change in the terminology of the reproductive structures. The structures which were formerly called "gametangia" should, therefore, be considered as "sporangia."

The principal method of reproduction in *Codium* is sexual. However, reproduction by fragmentation of the thallus is no doubt possible; but, since some time is required for fragments to produce rhizoidal anchorage organs, this method can hardly be of much importance, since fragments seldom remain stationary in moving water long enough to become attached. Asexual reproduction by detachable propagules which appear on the utricles and are either modified branches or immature sporangia have been reported in some species (Schmidt, 1923;

Fritsch, 1935). In the Beaufort plants of *C. decorticatum* such propagules are common in the spring and replace sexual reproduction at this time (Text Fig. 2). However, in summer, propagules have never been encountered, although hundreds of specimens from a variety of localities have been examined. Apparently propagules develop only during periods of low temperature when sporangia do not reach maturity. Functional gametangia are produced during summer and fall when water temperatures are high.

Periodic examination of sporangia indicates that development and discharge of gametes in *C. decorticatum* in the Beaufort estuary is directly correlated with tides. Sporangia may be found at all times in summer, but mature gametangia (i.e. sporangia which have become gametangia) are found only monthly at periods approaching spring tides. Gametes are discharged over a period of two or three days. Discharge takes place by day or night after low of spring tide. Plants brought into the laboratory during the period when gametes are being discharged can be separated into two groups: those with mature gametangia dark golden, or male plants; and those with mature gametangia dark green, or female plants. Since no plants have been observed with both kinds of gametangia it must be concluded that *C. decorticatum* at Beaufort is dioecious. Gamete discharge takes place through the tip of the gametangium in a manner similar to that described by Smith (1938) for *C. fragile* (Suring.) Hariot. A terminal plug of gelatin is shot out by an explosive force and the gametes move out through an apical pore of the gametangium and through a duct in the middle of the gelatin mass usually in single file. While passing through this pore and canal the gametes become much attenuated, but as soon as they are discharged they resume their normal shape. Gametes accumulate above the extruded mass of gelatin after discharge and remain stationary for about 60 seconds before becoming motile. Mrs. Longest (1946), of the University of North Carolina, using the Löffler staining technique on living material from this investigation, found that the two flagella of the microgamete of this species are of the same type as those reported for other green algae. Discharge of gametes in *C. fragile* takes place when plants are flooded after a period of intertidal exposure (Smith, 1938). *Codium decorticatum*, however, is not an intertidal species at Beaufort, but the tips of some of the plants are exposed during extremely low tides (Plate 9, fig. 1). Consequently, in *C. decorticatum* at Beaufort exposure is probably not a factor with which gamete discharge can be correlated.

From one to three sporangia are borne on each utricle, and sporangia of different ages may occur on the same utricle. There seems to be little or no localization on the plants in the production of sporangia, except that they are usually more numerous just below the branches. Microsporangia and macrosporangia in the Beaufort material of *C. decorticatum* are of equal shape and size (Text Fig. 3). According to Tilden (1935), in reporting on the cytological work of Williams (1925), the microsporangia are smaller than the macrosporangia in *C. dichotomum*. This is questionable since it is evident from the illustrations in Williams' paper that she worked with *C. fragile* instead of *C. dichotomum*, as was pointed out by Schmidt (1923). Went (1889) states that in monoecious species

the microsporangia appear about the time the macrosporangia are mature. Schmidt, however, reports that they develop simultaneously. In the Beaufort plants there seems to be a parallel development, and well developed sporangia may be found at any time throughout the summer, but microgametangia cannot be distinguished from macrogametangia until they are ripe and about ready for discharge.

GROWTH

Growth in *Codium* takes place by the addition of new utricles and by their enlargement. These are formed at the tips (apical growth) and also between preexisting utricles (intercalary growth). From the base of each utricle several branches arise. Some grow into the "medulla," while some grow lengthwise between the "cortex" and the "medulla," and sooner or later curve outward and become dilated to form the new utricles.

Inasmuch as mature plants differ in general appearance from young plants an experiment was made to determine how much effect intercalary growth has on the general form of the thallus. Several young plants (typical *C. dichotomum* forms) were used in this experiment. Strings were tied around the fronds one centimeter apart from the place of attachment to the tips. After a month of growth (July 14 to August 15) the plants gave some interesting results. The average plant showed an increase in length of 37 per cent. Growth took place throughout the length of the plant with considerably more growth in the upper portions of the thallus. In most specimens the upper fifth of the thallus grew as much as the remainder of the plant, with considerable apical growth. In 32 days of growth, 90 tips of the *C. dichotomum* form grew from 3-5 to 10-18 cm. in length per plant; while 90 large tips of the *C. decorticatedum* form (except those that were eaten back by natural enemies) grew from 5-10 to 20-28 cm. in length, and showed considerable flattening. From the above evidence one would conclude that intercalary growth is largely responsible for the change of a plant from the *C. dichotomum* form to the large flattened form characteristic of *C. decorticatedum*.

Regeneration from broken ends of the thallus is frequent and produces numerous small branches. Twelve large plants of the *C. decorticatedum* form were trimmed back to within an inch of the shell on which they were attached. After three weeks of growth many small branches were regenerated, each of which grew into the common juvenile *C. dichotomum* form.

DISTRIBUTION

Codium decorticatedum has a wide general distribution. According to Hoyt (1920) it is found from North Carolina to Florida; West Indies; Lower California, Mexico; South America; Europe; and Africa. As far as is known it reaches its northern limit of distribution on the eastern coast of North America at Ocracoke, North Carolina, where it was collected in the summer of 1946. It is present the year around in the Beaufort estuary, on the jetty at Cape Lookout, and on the seawall at Fort Fisher. However, the plants begin to diminish in size in the

middle of fall and by winter are represented by small stubs or cushions which could easily be overlooked if they were not marked for observation on the substrata on which they are attached.

Apparently temperature is the principal factor determining the northern distribution of *C. decorticatedum* on the east coast of the United States. Charts of thermograph records show that warm masses of water are abruptly broken off from Cape Hatteras (Church, 1936) where the annual range of temperature is 20 degrees C. The annual range of temperature of the sea at Beaufort is also 20 degrees C., but the summer temperature is warmer than the coastal area north of Cape Hatteras. As far as is known, North Carolina is the northern limit of range for many tropical algae such as *Codium*, *Dictyota*, and *Padina*. By more thorough collecting some of these species may be found at points north of North Carolina, but this is unlikely, because these stenothermal tropical species may not be able to persist under the eurythermal conditions found in areas north of North Carolina. According to Setchell (1915), temperature plays a very important part in the distribution of marine algae.

A study of the physical factors in the general distribution of *C. decorticatedum* along the east coast of the United States indicates that salinity is not a limiting factor in its northern distribution, since the same salinity prevails further north where *Codium* has not been reported. However, a study of the local distribution of this species in the Beaufort estuary shows that it disappears in places where salinity is less than half of sea water for periods of two or more days.

Large natural beds of *Codium* found at Beaufort are located on clear sandy shoals (Plate 9, fig. 1). However, scattered plants may be found anywhere they can find suitable places for attachment. They are also abundant on artificial breakwaters. Where they grow on shoals they do not grow in sand but are always attached to parts of shells which project out of the sand. The two most common of these shells furnishing attachment here are *Noetia ponderosa* Say and the pinna shell (*Atrina serrata* Sowerby). *Codium* grows on both the living and dead of these two species. Plants have been found growing on a great variety of objects such as rubber hose, tiling, bricks, concrete, communication cables, wire, and shells of Mollusks. However, two factors seem to determine whether *Codium* will become attached on a given object. The object must be within the range of the vertical distribution of *Codium*, and it must present a rough surface. Smooth, firm objects free of any growth of *Codium* are frequently found in beds of *Codium*. Most oyster shells do not furnish suitable places of attachment.

In order to determine the time necessary for *Codium* to settle and grow to visible proportions, rocks were placed in natural beds of *Codium*. After four weeks, some of the rocks showed small, upright, macroscopic growths. The growth might have been more pronounced if the rocks had been set out just before spring tide, since zygotes appear to be formed monthly just after spring tide.

Codium was never found growing more than 25 inches below mean low tide level in the Beaufort estuary, but at Cape Lookout, where the water is clearer, it was found growing four or more feet below mean low tide level. In the

Beaufort estuary it is strictly a sublittoral species, but on the jetty at Cape Look-out, on the exposed coast, it sometimes occurs in the lower littoral zone during periods of continuous wetting from wave action.

In certain habitats the vertical distribution may be correlated with light factors. Inasmuch as *Codium* is an attached plant it does not show periodic vertical migration as do many plankton. Consequently, light becomes a limiting factor below depths where its quantity and quality are insufficient for the minimum requirements from photosynthesis. The presence of suspensoids and dark material in solution in the water of the Beaufort estuary is variable in time and from place to place. Low photometer readings were correlated with poor or no growth of *Codium* in places where suitable attachment was available.

In the Beaufort vicinity certain sandy shoals offer the best places for the growth of *Codium*. Here the water is clear and light penetration is equivalent to that found in any part of the inlet. Currents in these places do not bring mud or silt, and light penetration is decidedly better. Stunted plants grow in places of low light intensity and they are generally darker green than those which grow under strong light intensity. Plants with a light green color placed in the tanks of running marine water in reduced illumination of the laboratory become dark green in about 18 hours. Observation of the utricle of these plants shows that their chloroplasts have migrated to the apices of the utricles. This would indicate that *Codium* has a mechanism for adjusting itself to changing conditions of light intensity within certain limits. The chloroplasts are dispersed under strong intensities and are concentrated near the surface of the thallus under low intensities.

NATURAL ENEMIES

Of the animals that were observed to feed on the thallus of *Codium*, the "sea hare" (*Tethys protea* Rang.) was the most destructive. Several of these animals were brought into tanks in the laboratory in order to determine approximately how much *Codium* they can consume. Over night one "hare" ate three medium sized plants. Diamondback terrapins, in the breeding pens at the Bureau of Fisheries, Beaufort, North Carolina, consume large quantities of *Codium* and some fishes in marine aquaria there eat it in lesser quantities. Although not as destructive as the "sea hare," a spider crab (*Libinia dubia* Milen-Edwards) and a mollusk (*Fasciolaria tulipa* L.) were also observed to feed on *Codium*. On several occasions the natural beds of this plant showed indications of excessive grazing, which might indicate that some unobserved swimming animals such as fish also feed on *Codium*.

Grateful acknowledgment is here given to Dr. H. L. Blomquist, Department of Botany, Duke University, who directed much of this work.

SUMMARY

1. The two forms of *Codium* at Beaufort, N. C., which have been referred by earlier investigators to *C. dichotomum* and *C. decorticatum* respectively must be

assigned to the latter species upon the following bases: (a) size of utricles, (b) mature form, (c) both forms occurring on the same thallus, and (d) the experimentally-induced growth forms.

2. From the material examined *C. decorticatum* at Beaufort is dioecious.

3. Growth is both terminal and intercalary and amounts to about 37 per cent during one mid-summer month.

4. There is strong evidence that discharge of gametes is periodic in relation to monthly spring tides.

5. The limit of the northern distribution of *Codium* is determined by a sudden drop in temperature of sea water at most seasons at Cape Hatteras, N. C.

6. Local distribution is determined by the following factors: (a) suitable objects of attachment within its vertical range, (b) degree of turbidity as it affects suspensoids and light penetration, and (c) salinity.

7. Several animals have been observed to be natural enemies of *Codium*.

8. *C. isthmocladum* Vickers is reported for the first time as growing attached in North Carolina waters.

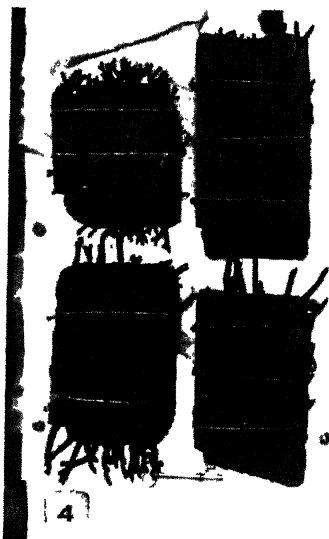
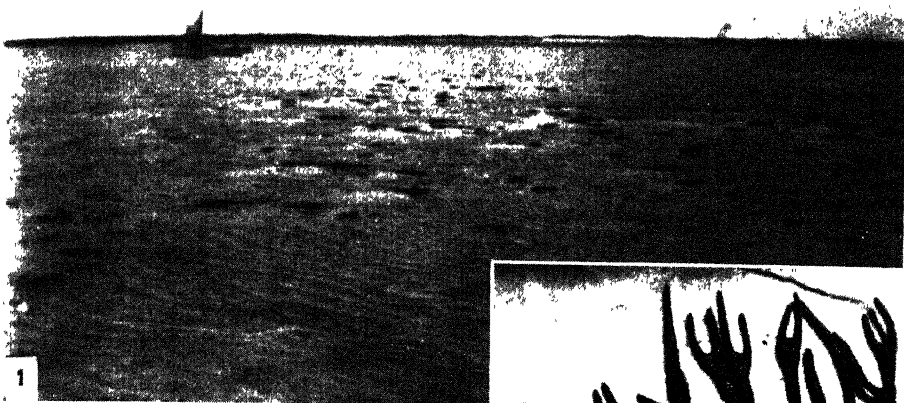
LITERATURE CITED

- CHURCH, PHIL E. 1936. Temperatures of the Western North Atlantic from Thermograph Records. U. of Wash. and Woods Hole Ocean. Contrib. No. 121.
- FRITSCH, F. E. 1935. The structure and reproduction of algae. Vol. 1. New York. Macmillan Co.
- HOYT, W. D. 1920. Marine algae of Beaufort, N. C., and adjacent regions. Bull. (U. S.) Bur. Fisheries 36: 372-550.
- LONGEST, PAULINE M. 1946. Structure of the cilia in *Ectocarpus mitchellae* and *Codium decorticatum*. Jour. E. Mitch. Sci. Soc. 62: 249-252.
- OLTMANN, D. F. 1904. Morphologie und Biologie der Algen. Jena, Verlag von Gustav Fischer.
- SETCHELL, W. A. 1915. The law of temperature connected with the distribution of marine algae. Ann. Missouri Bot. Gard. 2: 287-305.
- SCHMIDT, O. C. 1923. Beiträge zur Kenntnis der *Codium* Stack. Bibliotheca Bot. 23: Heft 91: 1-68.
- SMITH, G. M. 1938. Cryptogamic Botany. Vol. 1. New York, McGraw-Hill Book Co.
- TAYLOR, W. R. 1928. The marine algae of Florida and Dry Tortugas. Papers from the Tortugas Lab. of the Carnegie Inst. of Washington 25: 1-219.
- THURET, G. 1850. Ann. Sci. Nat. Bot. 3rd ser. 14: 214-260.
- TILDEN, J. E. 1935. The algae and their life relations. U. of Minnesota Press, Minneapolis, Minn.
- WILLIAMS, M. M. 1925. Contributions to the cytology and phylogeny of the Siphonaceous algae. I. The cytology of the gametangium of *Codium tomentosum* (Stackh.) Proc. Linnean Soc. New South Wales 50: 98-111.

PLATE 9

- Figure 1. Natural bed of *Codium decorticans* on shallow sand flats located west of Piver's Island in the Beaufort estuary. This photograph was taken at low water mark of the lowest tide of the summer. Normally only the tips of some of the plants are exposed during low of spring tide.
- Figure 2. *Codium decorticans* showing contrasting extreme forms common in the vicinity of Beaufort, North Carolina. The large, flattened form (upper) exhibits the habit of this species. The terete form (lower) has the external habit of *C. dichotomum* and has been incorrectly assigned to this species by phycologists who have worked with the material of *Codium* from this vicinity.
- Figure 3. Tips of *Codium*. (above) Densely branched tips (3-5 cm. in length), the *C. dichotomum* form of *C. decorticans*. (below) Broad, sparingly branched tips (5-10 cm. long) of the flattened form of *C. decorticans*. Ninety of each of the tips of these two forms were respectively attached to bricks (fig. 4) and grown under favorable conditions to produce flattened forms (fig. 5) of typical *C. decorticans*.
- Figure 4. Tips of *Codium* attached to bricks by string as they appeared at the beginning of 32 days of growth under identical and favorable environmental conditions. (above) the *C. dichotomum* form of *C. decorticans*. (below) *C. decorticans*.
- Figure 5. Tips of *Codium* after 32 days of growth. (left) The tips of the former *C. dichotomum* form. (right) *C. decorticans*.

PLATE 9



A CYTOLOGICAL STUDY OF THE EFFECTS OF COLCHICINE,
INDOLE-3-ACETIC ACID, POTASSIUM CYANIDE, AND
2,4-D ON PLANT CELLS

BY ALICE G. RYLAND

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina*

PLATES 10-14

This laboratory has been interested for some time in the nature and function of mitochondria in plants. It was hoped that by studying the cytological effects of certain poisons upon plant cells their pathology might yield some evidence as to the function of mitochondria. At least such a study should produce information about the physical changes of protoplasm preceding death, these changes to be expected in both the cytoplasm and the nucleus, which latter body is assumed to direct the ontogeny and metabolism of the cell.

Little is known as to the precise effect of poisons upon protoplasm, or, how chemicals kill, although the physiological reactions of many organisms to various poisons have long been studied. General statements to the effect that the protoplasm of cells when dead is coagulated, precipitated, or lysed give no clues to the specific actions of poisons on the components of living cells. For this reason it was decided to investigate the effect on plant cells of the four chemicals: colchicine, indole-3-acetic acid, potassium cyanide, and 2,4-D, which were chosen partly on account of their availability and common use in botanical research and also because, while all are poisonous in sufficiently high concentrations, their specific modes of action and the responses of plant tissues to them are all different.

The alkaloid colchicine has been used since 1937 as an agent for the production of polyploid plants (Blakeslee and Avery, 1937). It has also been used successfully in killing or inhibiting the growth of plant tumors (Bergner et al., 1940; Dermen and Brown, 1940; Havas, 1937).

Indole-3-acetic acid is a plant growth substance or auxin (Went, 1945), presumably a product of the metabolism of plant cells, having been isolated from them; but when used in other than extremely dilute concentrations it causes a marked growth response, gall or tumor formation, and eventual death of the affected cells. This growth response is used to practical advantage in producing roots on plant cuttings when the concentration is not high enough to be lethal, in producing parthenocarpic fruits, and in preventing fruit-drop.

Potassium cyanide is a powerful respiratory depressant even in very dilute concentrations and causes death as a result of anoxia (James and Hora, 1940).

As for 2,4-D, nothing is known of its physiological effect on plant cells due to the recency of its discovery as an herbicide. It is, however, one of the most effective weed-killers in use today (Grigsby, 1946).

Since the emphasis in this study was to be on the reaction of both mitochondria and chromosomes to the four poisons, much time was spent in preliminary experiments attempting to discover a fixing fluid that would preserve equally well the cytoplasmic and the nuclear constituents of the cell. Heretofore mitochondria have been fixed best by a basic fluid and chromosomes by an acid one. Recent work by Newcomer (1946), indicates that a single fixative adequate for all cell details might be found. The present attempts, however, were unsuccessful, due to some extent perhaps to the variation in permeability of dividing and nondividing cells (Stern, 1946).

MATERIALS AND METHODS

All experiments were carried out on young root tips of *Allium* and *Narcissus* which were grown in Shive's nutrient solution (Miller, 1938) and the bulbs then transferred to the experimental solutions. For colchicine, a 0.2% (0.005M) concentration in tap water was used and root tips cut after exposures of 13, 25, 37, 49, and 61 hours. For the other reagents, all except the 0.05M KCN made up in Shive's nutrient solution, concentrations and times of exposure were as follows:

KCN	0.0065%—25 hours—0.001M
	0.0325%—25 hours—0.005M
	0.325% —25 hours—0.05M
2,4-D	0.01% —25 hours
	0.05% —13 hours
	0.1% —25 hours
Indole-3-acetic acid	0.001% —25 hours

In the monocotyledonous plants used there is a periodicity of cell division which reaches its peak at 11 o'clock, morning and night. In order that advantage could be taken of this maximum division period and a large number of cells be obtained in stages of mitosis, bulbs were put into the reagents at 9 or 10 o'clock, left in for one or more 12-hour periods, and then root tips cut at a subsequent 11 o'clock hour. This schedule was followed for all cuttings.

Standard fixation fluids were used: Zirkle's (Johansen, 1940) for a basic image to show mitochondria and modified Chicago (Chamberlain, 1932) for chromosomes and spindle fibers. The latter was found to cause excessive shrinkage in the root tip cells so was replaced for most of the experiments by weak Flemming (Chamberlain, 1932), which preserved the spindle and chromosomes without distortion of the tissues. Zirkle's fluid was adjusted with the aid of a Macbeth potentiometer to a pH of from 4.4 to 4.8 using 10% KOH, NaOH, and NH_4OH , the last named seeming to give the best results.

Fixation was for 48 hours; then the tissue was washed for 8 to 12 hours, dehydrated in a series of ethyl-tertiary butyl alcohols, embedded in paraffin, and sectioned at 5 microns. Before staining in Heidenhain's hematoxylin, with or without counterstain of Orange G in clove oil, the slides were bleached in 1% KMnO_4 and 2.5% oxalic acid and mordanted overnight in 4% iron alum. Lang's (1936) modification of Heidenhain's iron alum, i.e., iron alum acidified with sul-

furic and acetic acids, was found to be superior in keeping properties and intensity of staining to the pure iron alum solution. Clarite was used as the mounting medium.

For purposes of comparison or control, normal root tips were cut at an eleven o'clock hour from bulbs grown in Shive's nutrient solution, fixed in both Zirkle's and weak Flemming killing fluids, dehydrated, embedded, sectioned, and stained as were the experimental tissues.

All pictures were taken with a Leitz "Makam" camera at an original magnification of 900 \times .

OBSERVATIONS AND DISCUSSION

To avoid repetition, all discussion of chromosomes will be assumed to refer to weak Flemming or Chicago fixations and discussion of mitochondria, vacuoles, or cytoplasm will refer to tissues fixed with Zirkle's fluid. For the specific fixation, hours of exposure, concentration of poison, and plant material used for each illustrative figure, see explanation of plates.

A. *Colchicine*:

The appearance of meristematic cells while under the influence of colchicine is so striking that the term "c-mitosis" has been coined to describe it. The mechanism of this c-mitosis has been investigated and described by Levan, Dermen, and many other writers (see Dermen, 1940, for review of subject). Colchicine apparently lowers the viscosity of the cytoplasm to a point at which spindle fibers cannot be formed (Beams and King, 1938). Consequently, the normal process of chromosome division is halted at metaphase and the sister chromatids appear much contracted, uncoiled, and held together only at the kinetochore, the pairs scattered in a diakinesis-like manner within the cell (Levan, 1938). Eventually this kinetochore connection breaks and as a result the cell now has twice the usual number of chromosomes. If allowed to recover from the drug, this cell will reconstruct its nucleus and on succeeding divisions maintain its tetraploid, or $4n$, state. Repetition of this chromosome division unaccompanied by cell division may result in octaploid, 16-ploid, and even 128-ploid cells (Walker, 1938); but cells with high chromosome numbers seldom persist, as such nuclei with difficulty become reconstituted and resume normal mitosis after removal of the drug (Beams and King, 1938). Chromosome doubling will continue as long as the cells are exposed to colchicine of the required concentration. In concentrations above 0.4% (Hawkes, 1942) colchicine is extremely toxic and causes death; also at lower concentrations, when exposure is prolonged and excessive chromosome doubling has occurred, cells are unable to recover. The effectiveness of colchicine in killing plant tumors is thought to be due to a more rapid response of tumor cells than normal cells to the drug and, consequently, excessive polyploidy leading to degeneration in that tissue (Brown, 1939; Dermen and Brown, 1940).

In the present study typical c-mitoses were observed (Figs. 16 and 17). Spindles were lacking and lobed and apparently polyploid nuclei were abundant.

in the tissues exposed for 25 hours (Fig. 16). There was not a great deal of division in the tissues given longer treatment; this fact appears to corroborate Levine and Gelber's (1943) observation that the greatest number of metaphase stages occurs at 25 hours after beginning of treatment of the roots with colchicine solution.

Since colchicine is assumed to lower the viscosity or surface tension of the cytoplasm, as is demonstrated by inhibition of spindle formation, its effect on the mitochondria should show whether or not they, too, are sensitive to such changes. Meristem mitochondria were short and rod-like but differed from those of the controls in the tendency to constriction and fragmentation. This was more pronounced in the exposures of 25 hours or longer (Figs. 8 and 9) than in the 13 hour stage (Fig. 7). There was no polymorphism of mitochondria in the older tissues of the root and with few exceptions, regardless of the length of the exposure, they had rounded up into ball-like bodies, some apparently coalescing (Figs. 11-15). Fragmentation was observed in the region of elongation (Fig. 10) after 37 hours of exposure. These activities of the mitochondria: rounding up, coagulating of small globules into larger ones, fragmenting of rods into smaller, rounder bodies, and shortening and thickening of other rods, all indicate a response to lowered surface tension and suggest that mitochondria are sensitive to the viscosity of the surrounding medium.

A secondary effect of colchicine is the swelling occurring in the region of elongation of the young root. These swellings appeared as expected on the roots treated in these experiments. They are not due to polyploidy of the cells, as was once thought; the number of cell rows also is not increased. Each cell, however, seems to have increased in width, thus causing a bulging of the root just above the meristem. According to Hawkes (1942), these cells have roughly the same volume as do cells in a corresponding region in untreated roots, growth occurring in all directions instead of mainly longitudinally.

B. *Indole-3-acetic acid*:

That the concentration of the indole-3-acetic acid used was not strong enough to prevent further growth was demonstrated by replacing the bulb in Shive's nutrient solution after exposure to the reagent and observing the resumption of normal root growth beyond the swollen area. Sub-terminal root swellings are a characteristic result of treatment with indole-3-acetic acid and some other growth substances as well as with colchicine (Levan, 1939). Swellings were conspicuous after 24 hours of exposure to the 0.001% solution. As in the case of the colchicine treatment, this bulging at the region of elongation is not due to an increase in the number of cell rows but to a widening of individual cells of this region. These swellings, strangely enough, do not occur with either colchicine or indole-3-acetic acid treatment if the root tip has been removed (Hawkes, 1942) so are thought to be related in some way to the minute amount of auxin produced naturally in the root tip.

The most startling observation revealed in the study of these tissues was the unusual amount of cell division present, nearly every cell in the meristem being

in some stage of mitosis. Anaphase and telophase stages had normal spindles (Figs. 24 and 25) in contrast to the condition produced by colchicine. These experiments indicate that indole-3-acetic acid when applied from outside the tissue causes a stimulation of cell division that occurs at a slower rate under the regulation, we presume, of the plant's own auxin (Went, 1945). This technic should be useful in treating root tips for chromosome study.

Increased cell division is assumed to indicate increased metabolic activity. If mitochondria are involved in metabolism, they might be expected to become more numerous under this condition. The study of these cells, however, seemed to indicate a general reduction of the mitochondriome in the meristem as compared with that of the control tissues. Occasionally well-fixed cells showed swollen mitochondria and rod-shaped ones that were constricted as if fragmenting (Fig. 18). At the root-swelling above the meristem the mitochondria were mostly vesicular and often clumped (Figs. 21 and 23). The presence of vesiculated mitochondria in this area is not understood but may indicate that these cells have an increased permeability. Farther back in the region of maturation more clumping was observed, with polymorphic structures ranging from attenuated, branched, crooked, and sometimes vesiculated threads to the globular forms (Figs. 19, 20, and 22). The mitochondria, therefore, do not appear to increase with increased cell metabolism and their irregularities resulting from indole-3-acetic acid treatment are not explained by the present study.

C. KCN:

A number of workers have investigated the relation of KCN to oxygen consumption in various plants. James and Hora (1940) report that 0.002M KCN reduces respiration in barley leaves about 36%, not, however, preventing recovery in water. At a 0.02M concentration inhibition of oxygen consumption is complete and irreversible. Seifriz (1943) states that 0.01M KCN suppresses respiration 80% in *Physarum* yet does not stop protoplasmic flow so is not strong enough to cause death. The present experiments indicate that a concentration of 0.005M on root tissues of *Allium* does cause death of some cells and that at 0.05M necrosis is extensive. Necrotic cells were observed as first occurring in the meristem of the 0.005M concentration; the plerome appeared to be most resistant to the poison.

Both nuclei and cytoplasm of the tissues exposed to the strongest concentration were extremely difficult to stain. Prolonged mordanting with iron alum did not increase their stainability, nor did a four-day premordanting in $K_2Cr_2O_7$. In the meristem the chromosomes were extremely attenuated and ragged (Figs. 27, 28, and 30), presenting a markedly abnormal appearance faintly reminiscent of the "lampbrush" chromosomes as described by Kodani (1941). The matrix had largely disappeared. Nuclei of resting cells were large and pale, the chromatin content appearing sharply outlined in irregular strands and masses in the absence of stained nuclear lymph (Figs. 28 and 30). Back of the meristem the cells were colorless.

In a study of barley seedlings deprived of oxygen by being grown in an atmos-

phere of nitrogen for four days, Steinitz (1943) reports continuation of mitosis but with such abnormalities as chromosome clumping, sticky anaphase bridges, and extrusion of chromatic material into the cytoplasm. These observations were not corroborated in the present study since mitotic stages could not be identified beyond metaphase and all were poorly stained. Whatever the relationship of the nucleus to cellular respiration, and this study offers no enlightenment upon it, it is interesting to note that Steinitz, in observing effects of anoxia upon the nucleus, did not observe failure of formation of the chromosome matrix such as results from KCN treatment. This indicates that this condition may be due to a secondary effect not associated with anoxia. It would be interesting to see if the gene string were not also affected by KCN if organisms so treated were allowed to recover.

Mitochondria in living cells were extremely abnormal and generally coagulated (Fig. 26), in some cases swollen and in others fragmented (Fig. 29) and almost totally destroyed. None of the typically normal, filamentous types were found. This coagulation and destruction of mitochondria may lend some support to the hypothesis that they are associated with cellular respiration. The reaction of mitochondria to other respiratory-inhibiting substances should be observed. Steinitz, unfortunately for this study, was concerned only with the chromosome and nuclear phenomena of the cell.

D. 2,4-D:

According to Grigsby (1946), a 0.01% concentration of 2,4-D will kill young poison ivy growth, 0.05% concentration stops pollen production in ragweed, and 0.1% kills ragweed.

The most noticeable effects of 2,4-D in the present study were excessive vacuolation of the cytoplasm in the meristem, a general inhibition of cell division, and the production of c-mitotic metaphases. Very few dividing cells were observed at the 0.05% concentration, and although in tissues exposed to the higher concentration a great number of prophases were seen, no stages beyond metaphase were found and the chromatin itself appeared granular and much disturbed. The metaphase chromosomes had a typical c-mitotic appearance, the chromatids partly separated and pairs scattered in the cell without an observable spindle (Fig. 32). Polyploidy, however, was not evident. Other chemicals, among them acenaphthene, bromonaphthalene (Levan and Ostergren, 1943), and podophyllin (Sullivan and Wechsler, 1947), have been reported to cause c-mitosis, also without causing c-tumors. Perhaps in sufficiently dilute concentration 2,4-D might also produce polyploidy.

Meristem mitochondria in the 0.05% concentration were irregularly swollen and crooked (Fig. 31). In the 0.1% concentration, farther back in the root tissues, they had an especially fragmented appearance (Fig. 33). Death of cells was not observed in concentrations below 0.05%. At this strength, though, some cells of the inner periblem had been killed. At the higher concentration necrosis was more general; the epidermis and upper plerome seemed to be most resistant.

A general description of the morphological appearance of the cell preceding death by 2,4-D would be as follows: the chromatin network of the resting nucleus granular instead of the usual anastomosing threads, the mitotic cycle stopped, cytoplasm excessively vacuolated, and the mitochondria disintegrating and presenting a lysed appearance. Since cellular structure is the basis of cellular function, death apparently results from a general collapse of the morphological constituents of the cell which precludes any adequate description of how 2,4-D kills.

SUMMARY

Colchicine treatment indicates that mitochondria are affected by changes in the viscosity of the protoplasm.

The effect of colchicine upon the nuclear cycle corroborates the findings of previous investigators.

0.001% indole-3-acetic acid causes a phenomenal increase in the number of mitoses in the root tip meristem and the technic should have value in chromosome studies.

The metabolic stimulus of indole-3-acetic acid as indicated by increased cell division does not increase the number of mitochondria per cell.

KCN causes marked diminution of the chromosome matrix resulting in ragged chromosomes and it is suggested that the gene string may be affected. Since this condition has not been reported in studies of respiratory inhibition using nitrogen, the observed effect may be due to causes other than anoxia.

KCN causes a marked loss of staining capacity of cells, which increases with the concentration.

KCN and 2,4-D cause a similar disintegration of the mitochondria preceding death of the cell. No conclusions can be drawn as to the specific cause of this condition.

2,4-D causes inhibition of cell division and the appearance of c-mitoses in the root tip meristem but no polyploidy. This latter condition might be obtained by treatment with a sufficiently dilute concentration of the poison.

This work was done under the direction of Dr. E. H. Newcomer, to whom I wish to express my gratitude.

LITERATURE CITED

- BEAMS, H. W., AND R. L. KING. 1938. An Experimental Study on Mitosis in the Somatic Cells of Wheat. *Biol. Bull.* 75: 189-207.
- BERGNER, A. DOROTHY, A. G. AVERY, AND A. F. BLAKESLEE. 1940. Chromosomal Deficiencies in *Datura stramonium* Induced by Colchicine Treatment. *Am. Jour. Bot.* 27: 676-683.
- BLAKESLEE, A. F., AND A. G. AVERY. 1937. Methods of Inducing Doubling of Chromosomes in Plants by Treatment with Colchicine. *Jour. Hered.* 28: 393-411.
- BROWN, NELLIE A. 1939. Colchicine in the Prevention, Inhibition, and Death of Plant Tumors. *Phytopathology* 29: 221-231.
- CHAMBERLAIN, C. J. 1932. *Methods in Plant Histology*. University of Chicago Press, Chicago.

- DERMEN, HAIG. 1940. Colchicine Polyploidy and Technique. *Bot. Rev.* **6**: 599-635.
- AND NELLIE A. BROWN. 1940. Cytological Basis of Killing Plant Tumors by Colchicine. *Jour. Hered.* **31**: 197-199.
- GRIGSBY, B. H. 1946. Some Effects of 2,4-D on Ragweed and Certain Woody Plants. *Mich. Agri. Exp. Sta. Quart. Bull.* **28**.
- HAVAS, LASZLO J. 1937. Colchicine, "Phytocarcinomata" and Plant Hormones. *Nature* **140**: 191-192.
- HAWKES, J. G. 1942. Some Effects of the Drug Colchicine on Cell Division. *Jour. Genetics* **44**: 11-22.
- JAMES, W. O., AND F. B. HORA. 1940. The Effect of Cyanide on the Respiration of Barley. *Annals of Botany* **4**: 107-118.
- JOHANSEN, D. A. 1940. *Plant Microtechnique*. McGraw-Hill, New York.
- KODANI, M. 1941. The Structure of the Salivary Gland Chromosomes of *Drosophila melanogaster*. *Jour. Hered.* **32**: 147-156.
- LANG, A. G. 1936. A Stable, High-Contrast Mordant for Hematoxylin Staining. *Stain Tech.* **11**: 149-151.
- LEVAN, ALBERT. 1938. The Effect of Colchicine on Root Mitoses in *Allium*. *Hereditas* **24**: 471-486.
- . 1939. Cytological Phenomena Connected with the Root Swellings Caused by Growth Substances. *Hereditas* **25**: 87-96.
- AND G. OSTERBERG. 1943. The Mechanism of C-mitotic Action — Observations on the Naphthalene Series. *Hereditas* **29**: 381-443.
- LEVINE, MICHAEL, AND SIDNEY GELBER. 1943. The Metaphase Stage in Colchicized Onion Root-tips. *Bull. Torrey Bot. Club* **70**: 175-181.
- MILLER, E. C. 1938. *Plant Physiology*. McGraw-Hill, New York.
- NEWCOMER, E. H. 1946. Concerning the Duality of the Mitochondria and the Validity of the Osmiophilic Platelets in Plants. *Am. Jour. Bot.* **33**: 684-695.
- SEMFRIE, WM. 1943. Protoplasmic Streaming. *Bot. Rev.* **9**: 49-123.
- STEINITZ, LOTTI M. 1943. The Effect of Lack of Oxygen on Mitosis in Barley. *Am. Jour. Bot.* **30**: 622-625.
- STERN, HERBERT. 1946. The Permeability of Cells Undergoing Nuclear Division. *Trans. Royal Soc. of Canada* **40**: 141-148.
- SULLIVAN, B. J., AND H. I. WECHSLER. 1947. The Cytological Effects of Podophyllin. *Science* **105**: 433.
- WALKER, RUTH I. 1938. The Effect of Colchicine on Somatic Cells of *Tradescantia paludosa*. *Jour. Arnold Arbor.* **19**: 158-162.
- WENT, F. W. 1945. Auxin, the Plant-growth Hormone, II. *Bot. Rev.* **11**: 487-496.

EXPLANATION OF PLATES 10-14

PLATE 10: Controls—root tip tissues grown in Shive's nutrient solution

- Figs. 1, 2. *Narcissus*; Zirkle fixation; meristem.
- Fig. 3. *Narcissus*; Zirkle fixation; region of maturation.
- Fig. 4. *Narcissus*; Zirkle fixation; region of elongation.
- Fig. 5. *Allium*; weak Flemming fixation; showing mitosis.
- Fig. 6. *Allium*; weak Flemming fixation; showing spindle fibers.

PLATE 11: Colchicine, 0.2% (0.005M) concentration; *Allium* root tips

- Fig. 7. 13 hours; Zirkle fixation; meristem.
- Fig. 8. 25 hours; Zirkle fixation; meristem.
- Fig. 9. 61 hours; Zirkle fixation; meristem.
- Fig. 10. 37 hours; Zirkle fixation; region of elongation.
- Fig. 11. 15 hours; Zirkle fixation; region of elongation.
- Fig. 12. 49 hours; Zirkle fixation; region of elongation.

PLATE 12: Colchicine (continued)

- Fig. 13. 61 hours; Zirkle fixation; region of maturation.
Fig. 14. 13 hours; Zirkle fixation; region of maturation.
Fig. 15. 37 hours; Zirkle fixation; region of maturation.
Fig. 16. 25 hours; weak Flemming fixation; showing lobed nucleus and c-mitotic chromosomes.
Fig. 17. 13 hours; weak Flemming fixation; showing c-mitotic chromosomes.

PLATE 13: Indole-3-acetic acid, 0.001% concentration; 25 hours; *Narcissus* root tips

- Fig. 18. Zirkle fixation; meristem.
Fig. 19. Zirkle fixation; region of maturation, periblem.
Figs. 20, 21. Zirkle fixation; region of elongation.
Fig. 22. Zirkle fixation; region of maturation.
Fig. 23. Zirkle fixation; region of elongation.
Figs. 24, 25. Chicago fixation; showing unusual amount of mitosis.

PLATE 14: Figs. 26-30, KCN; Figs. 31-33, 2,4-D; *Allium* root tips

- KCN; 0.05M concentration; 25 hours:
Fig. 26. Zirkle fixation; meristem.
Figs. 27, 28. Weak Flemming fixation; note "ragged" chromosomes.
Fig. 29. Zirkle fixation; region of elongation.
Fig. 30. Weak Flemming fixation; note "ragged" chromosomes.
2,4-D:
Fig. 31. 0.05% concentration; 13 hours; Zirkle fixation; meristem.
Fig. 32. 0.05% concentration; 13 hours; weak Flemming fixation; chromosomes resembling c-mitotic ones.
Fig. 33. 0.1% concentration; 25 hours; Zirkle fixation; region of elongation.

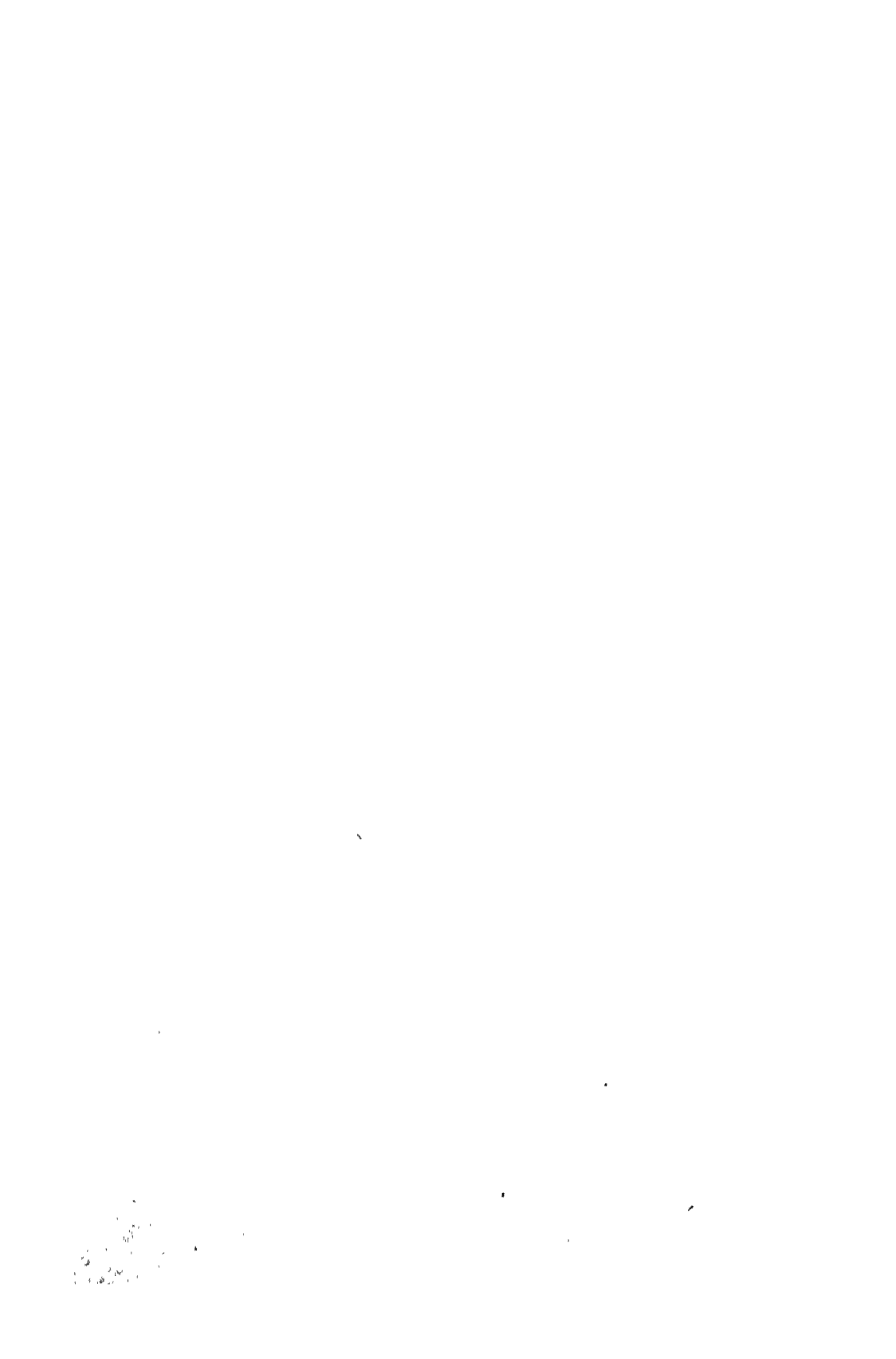


PLATE 10

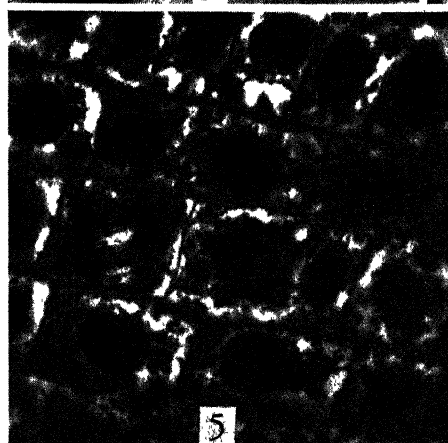
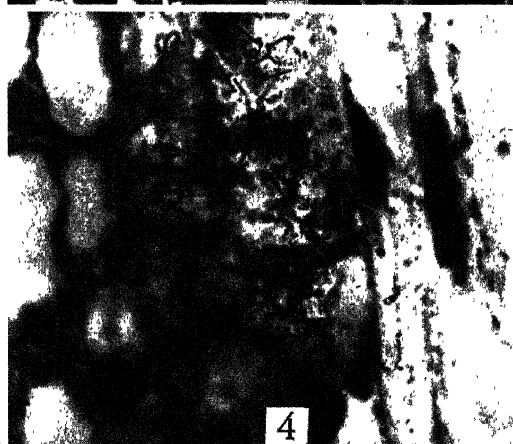


PLATE 11

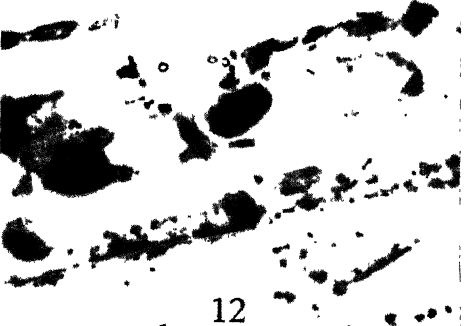
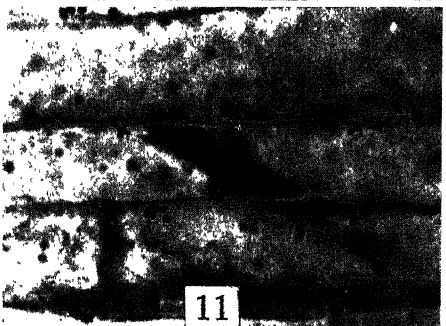
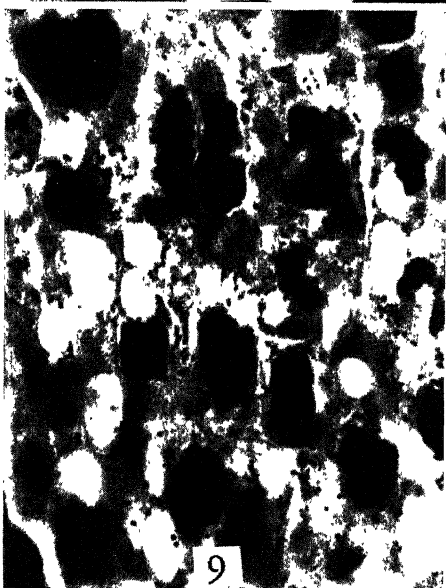
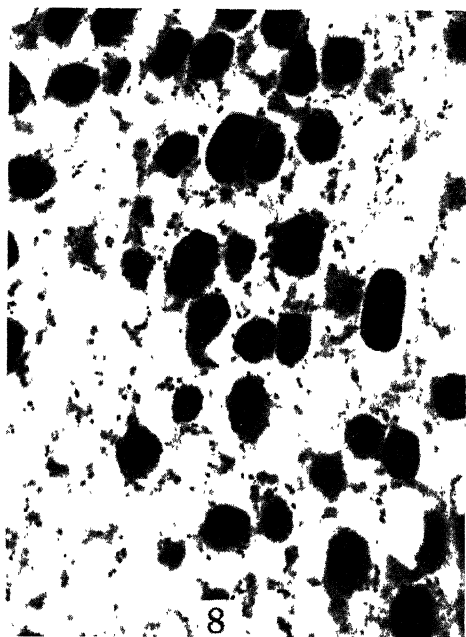


PLATE 12

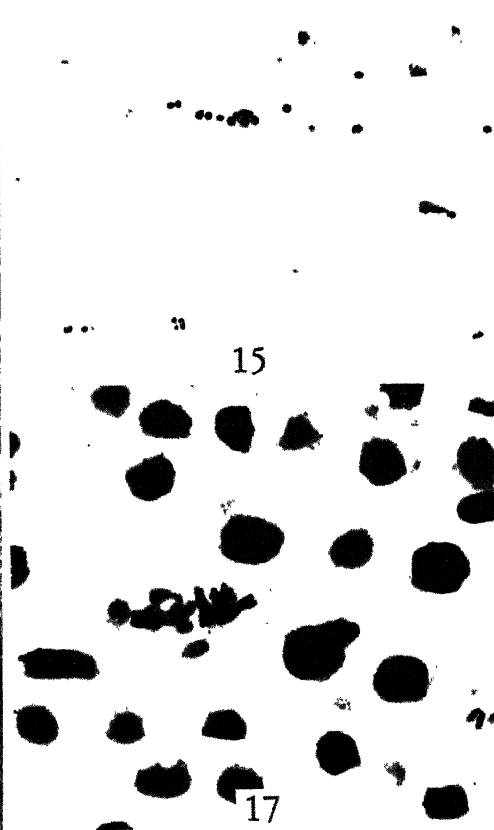


PLATE 13

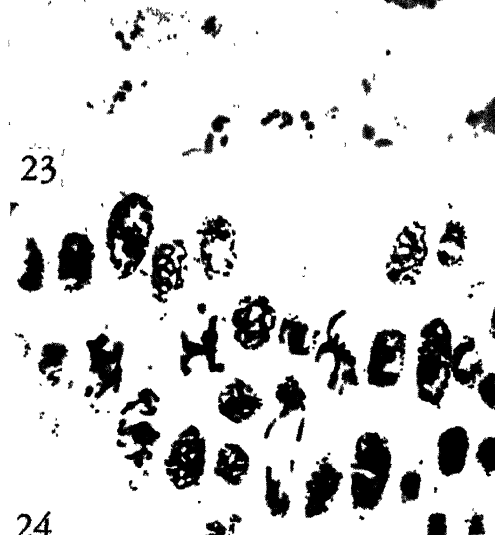
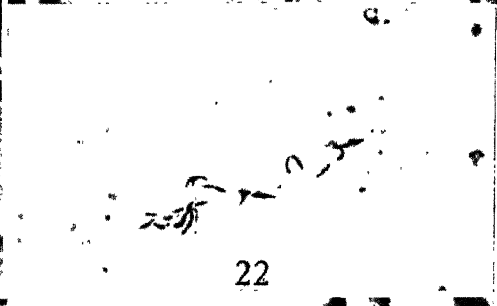
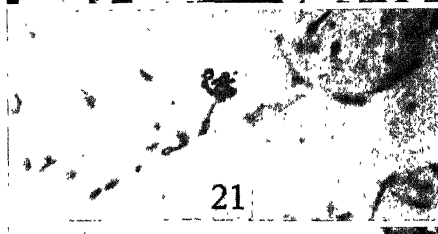
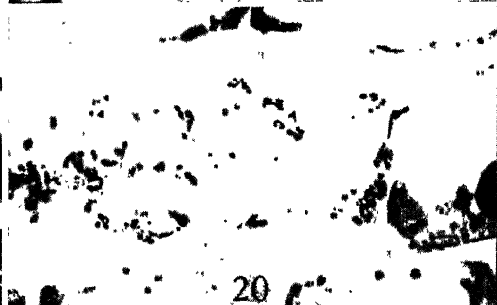
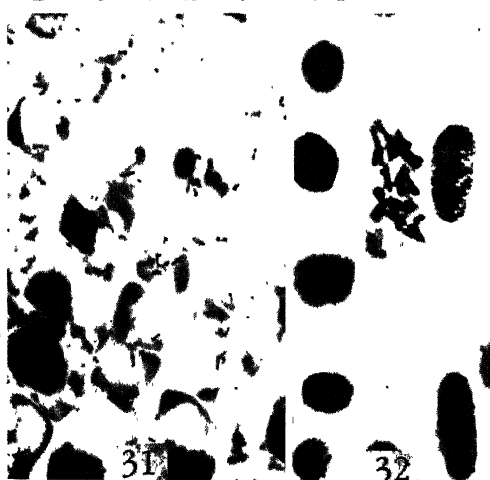
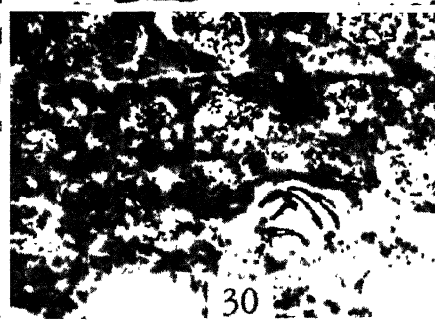
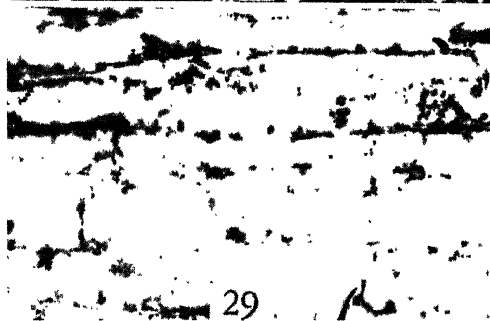
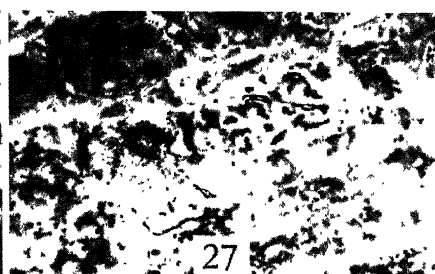


PLATE 14



A SECOND REPORT ON PARASITIC COPEPODS COLLECTED AT BEAUFORT, N. C.

BY A. S. PEARSE

Department of Zoology, Duke University
Durham, North Carolina

EIGHTEEN FIGURES

Through the courtesy of Dr. Reinard Harkema the writer received seven parasitic copepod crustaceans collected from fishes during August, 1947. These are in addition to those already reported from the Beaufort Region (Pearse, 1947). Two of them are new. The findings are described in the following systematic account. Grateful acknowledgement is made to Paul L. Illg, who compared the writer's specimens with those in the United States National Museum and made constructive suggestions.

Order COPEPODA

Suborder CALIGOIDA

Family CALIGIDAE

Caligus mutabilis Wilson 1905

A single female was taken from the channel bass, *Sciaenops ocellatus* (L.), on August 20.

Family EURYPHORIDAE

Paralebion elongatus Wilson 1911

Three females were taken from a sharp-nosed shark, *Scoliodon terraenovae* (Richardson), on August 20.

Family ANTHOSOMIDAE

Lernanthropus longipes Wilson 1932

Two females were found on the channel bass, *Sciaenops ocellatus* (L.), on August 20.

Family EUDACTYLINIDAE

Krøyeria spatulata n. sp.

Four females, two of which carried egg strings, were taken from a sharp-nosed shark, *Scoliodon terraenovae* (Richardson), on August 20.

Carapace one-fourth wider than long, with lateral margins rounded but angulate and also rounded at the posterior lateral angles; styliiform processes (Figs. 1, 6) reaching the anterior fourth of the fourth thoracic segment and bifid at the tip. Fourth thoracic segment a little longer than second and third,

which are about equal; fifth segment a little longer than any of the others and widened a little posteriorly. The fused fifth and genital segment is narrower than the thoracic segments, is a little more than three times as long as the head and thorax, and is nearly fifteen times as long as wide. Abdomen composed of a single segment, tapered posteriorly, one-seventh as long as fifth-genital segment. Caudal rami (Fig. 9) a little more than one-fourth the length of the abdomen; tipped with two plumose setae, a plain seta, two short spines, and two blunt tubercles.

First antenna (Fig. 2) 6-segmented, the first segment longest, the terminal segment tipped with four long and four short setae. Second antenna (Figs. 3, 4) short and stocky, the movable finger stout and moderately curved, the fixed finger broad, spatulate, rugose at tip. Second maxilla (Fig. 5) with basal segment short, second segment curved, and terminal claw about one-third as long as second segment. Maxilliped with basal segment straight and little swollen; terminal claw as long as basal segment but curved sharply just beyond the middle.

All the legs are biramose and the rami are 3-segmented. The terminal segment of the short endopod of the first leg (Fig. 7) is armed with a terminal, non-plumose seta and seven strong terminal and lateral spines. The segments of the first exopod bear 1, 1, and 3 plumose setae and the terminal segment has one non-plumose seta. The rami of the fourth leg (Fig. 8) are about equal in length. The endopod bears 1, 1, and 3 (short) plumose setae and 0, 3, and 5 medial spines; the exopod, 1, 1, and 5 plumose setae and a short terminal non-plumose seta and three small sub-terminal spines. The egg strings (Fig. 10) are linear; on one female each contains 27 eggs, on the other, 39. Length of type female, 6.0 mm.; of egg-bearing females, 7.5 mm.

Type: U. S. National Museum No. 84219.

Male, unknown.

This species is named for the unique spatulate character of the fixed arm of the chela of the second antenna. It somewhat resembles *Krøyeria papillipes* Wilson 1932, but differs from it in the character of the second antenna, the lack of papillae on the middle endopod segment of all legs, the armature of the legs and the caudal rami, and the length of the styliform process.

Nemesis atlantica Wilson 1922

Two females were collected from a sharp-nosed shark, *Scoliodon terraenovae* (Richardson), on August 20.

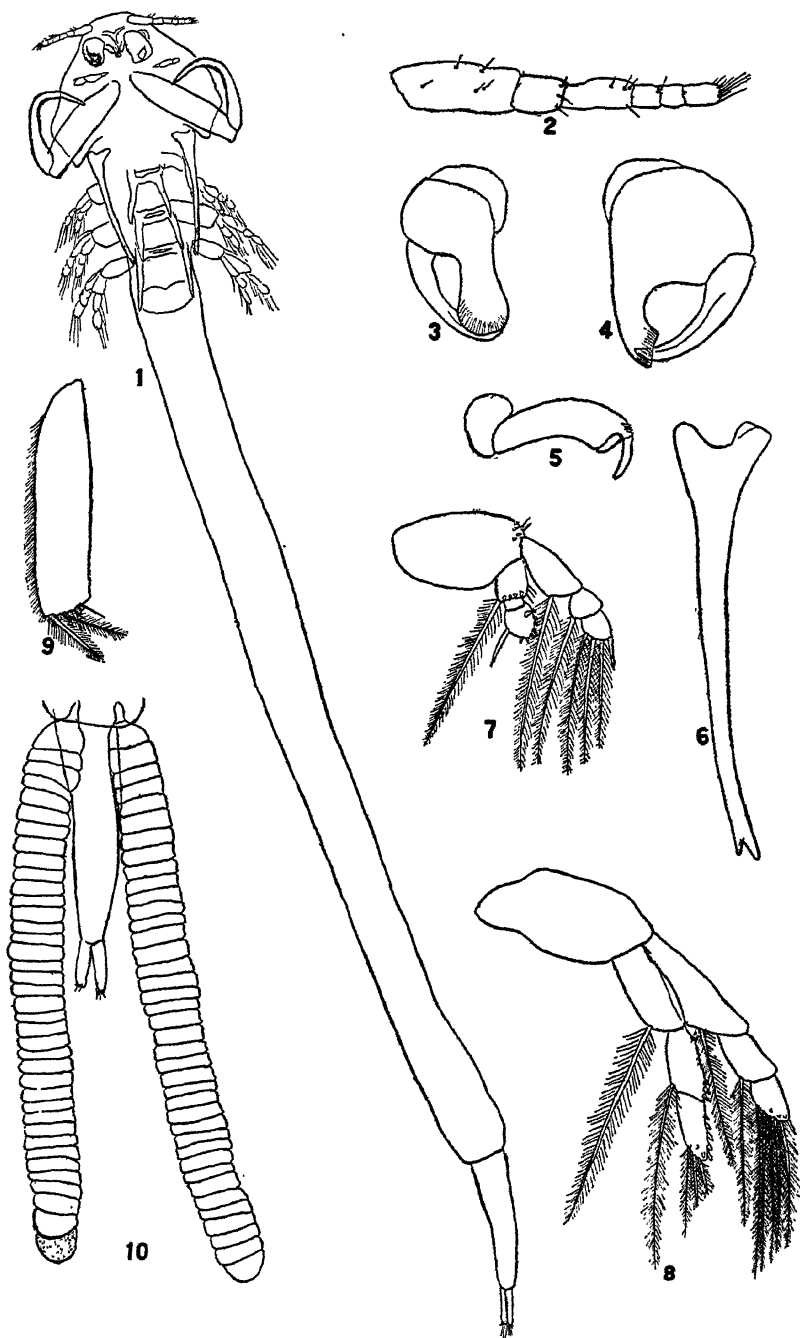
Eudactylinella alba Wilson 1932

Five females, one with eggs, were taken from a spotted whip ray, *Aetobatus narinari* (Euphrasen), on August 19.

Family DICHELESTHIIDAE

Hatschekia harkema n. sp.

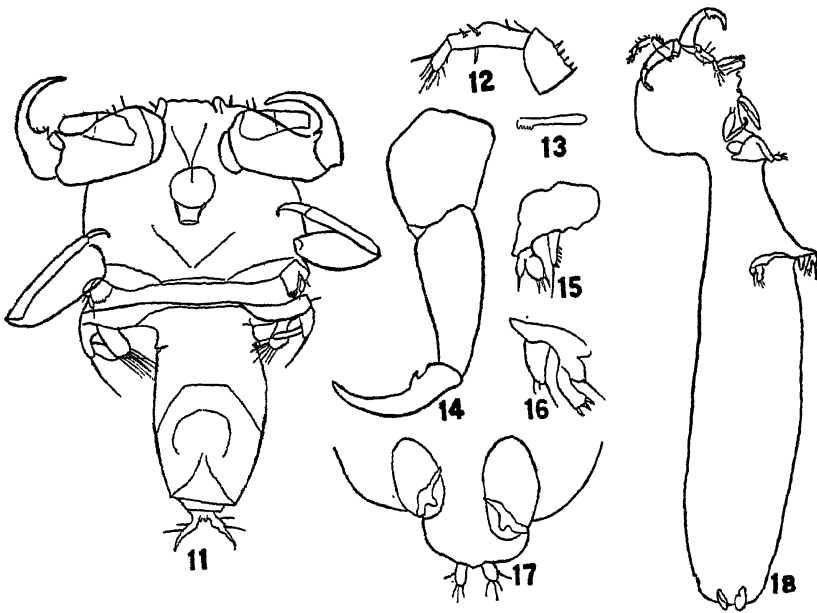
Five females and one male were collected from a spiny boxfish, *Chilomycterus schoepfi* (Walbaum), on August 13.



FIGS. 1-10. *Kroyeria spatulata* n. sp.: 1, female ventral view; 2, first antenna; 3, 4, second antennae; 5, maxilliped; 6, styliform process; 7, right first leg; 8, right fourth leg; 9, right caudal ramus; 10, abdomen and egg strings.

Female.—Head swollen dorsally and followed by a narrower neck (Fig. 18): the former is about 0.4 mm. wide, the latter 0.2 mm.; the linear body in its widest part is about 0.4 mm. wide; total length of head and body 1.8–2.4 mm. The anterior end of the body is more tapered than the posterior, which is gradually rounded. The abdomen is minute (0.04 mm. long; Fig. 17) and bears the caudal rami, which are 0.02 mm. long and each has three terminal and one lateral setae.

First antennae (Fig. 12) 3-segmented, the middle segment longest, the terminal segment shortest. Second antenna (Fig. 14) 3-segmented; the first segment about as wide as long; the second segment tapered somewhat distally, about twice as long as wide; terminal claw two-thirds as long as second segment, acute,



FIGS. 11–18. *Hatschekia harkema* n. sp.: 11, male, ventral view; 12, female first antenna; 13, mandible; 14, second antenna, female; 15, left first leg, female; 16, right second leg, female; 17, abdomen and caudal rami of female; 18, side view of female.

curved, bearing a recurved tooth on the inside near its base, and with a low tubercle at the base. The mandibles bear five teeth on their medial margin near their tips (Fig. 13). The maxilliped bears a prominent sharp tooth at its base; the first segment is less than half as long as the second; the second segment is equal in length with, but twice as wide as, the third; the terminal claw is less than one-third as long as the second segment. The first legs (Fig. 15) are biramous; the exopod is indistinctly 2-segmented, the basal lobe bears two terminal setae, the terminal lobe bears three spines; the inner lobe bears a terminal seta and five medial spines. The second leg (Fig. 16) is biramous; the exopod bears three terminal spines and a lateral seta; the endopod is tipped by three setae.

Male (Fig. 11).—Very minute, 0.4 mm. long. The head is covered by a dorsal plate which is rounded in front and truncate behind; this is slightly wider than long. The first two segments of the thorax are as wide as the head and clearly defined. The other thoracic segments and the genital segment are fused and are almost as long as the head and preceding thoracic segments. The abdomen is one-fifth as long as wide; it bears two long pointed appendages, each of which has two lateral setae.

The first antenna is short and truncate. The second antennae and maxillipeds are similar to those of the female but relatively larger and stronger. The first legs are biramose; the 2-segmented exopod bears a terminal seta, the endopod is rough at the tip. The second legs are also biramous and the exopod is 2-segmented; the exopod bears a lateral seta on the basal segment and a terminal seta on the slender distal segment; the endopod bears six terminal setae.

Types: U. S. National Museum No. 84217.

This species differs from Wilson's (1913) *H. oblonga* in the greater length of its thoracic segments, in having a strong spine at the base of the terminal claw of the second antenna, and the greater number of spines on the thoracic legs. In Wilson's (1922) key to the genus *Hatschekia* the species agrees with *H. linearis* in having 3-segmented antennae and yet disagrees in that the second thoracic segment is narrower than the head; the width of the head agrees with *H. hypoglossi* but does not agree in that the antennae are not 5-segmented. None of the females carried egg strings, so they may have been immature. The species is named for the collector, Dr. Reinard Harkema.

Four copepods that were previously reported (Pearse, 1947) were also collected by Dr. Harkema from the same hosts: *Ergasilus lizae* Krøyer and *Naobranchia lizae* (Krøyer) from *Mugil cephalus* L.; *Pseudocycnus buccatus* Wilson from *Scomberomorus maculatus* (Mitchill); and *Pseudochondracanthus diceraus* Wilson from *Sphaeroides maculatus* (Bloch & Schneider).

REFERENCES

PEARSE, A. S.

1947. Parasitic copepods from Beaufort, North Carolina. Jour. Elisha Mitch. Sci. Soc. **63**: 1-16.

WILSON, C. B.

1913. Crustacean parasites of West Indian fishes and land crabs. Proc. U. S. Nat. Mus. **44**: 189-277.
1922. North American parasitic copepods belonging to the family Dichelesthidae. Proc. U. S. Nat. Mus. **60**: 1-100.

A NEW MEMBER OF THE PLASMODIOPHORACEAE

BY WILLIAM R. PENDERGRASS

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina*

PLATE 15

A new species of *Octomyxa* parasitic in *Brevilegnia linearis* was found in a soil collection made by the writer from the City Park, Florence, S. C., in June 1947. The swollen ends of the *Brevilegnia* hyphae suggested that it might be *Octomyxa Achlyae* Couch, Leitner, and Whiffen (1939) and upon close examination the resting spores were seen to be in groups of eight. A study was begun to determine whether the spherical galls were due to *O. Achlyae* or to a new species.

Material was killed and fixed in Randolph's modified Navashin fluid (Johansen, 1940), embedded in paraffin, and sectioned at 10 microns. As a stain, Couch's (1932) modification of Gram's gentian violet technique was used. With this technique, the "protomitotic" type of nuclear division within the developing parasite plasmodium was demonstrated (Plate 15, fig. 3). This primitive type of nuclear division was also reported by Couch et al.

Using the Löffler flagella staining technique (Couch, 1941), zoospores were seen to have one short anterior flagellum and one long posterior flagellum. Biflagellate heterokont zoospores were found also in *O. Achlyae*. Ledingham (1934) reported biflagellate zoospores in *Plasmodiophora brassicae* and *Spongospora subterranea*, and zoospores with one long and one short flagellum in *Polyomyxa graminis* (1939).

Thus various characteristics considered as prerequisites of a member of the Plasmodiophoraceae by different workers, mainly hypertrophy of the host, "protomitotic" type of nuclear division, negative cellulose reaction, resting spores in a definite grouping, and biflagellate heterokont zoospores, are all fulfilled by this fungus.

At the suggestion of Dr. J. N. Couch, a comparison was made of the size of galls, zoosporangia, and resting spores of *O. Achlyae* with those of the fungus parasitic on *Brevilegnia*, and a distinct difference was found. The galls of *O. Achlyae* measured 50–150 μ , as compared to 30–90 μ for those of the new species; zoosporangia of the former 6–16 μ in thickness with 6–14 zoospores in each, zoospores of the latter 5.2–7.6 μ with 4–6 zoospores in each; however the individual resting spores in *O. Achlyae* were smaller in diameter (2.4–3.2 μ) than those of the parasite on *Brevilegnia* (2.8–4.9 μ).

The host range studies also indicated a difference. Numerous attempts were made to induce parasitism of this fungus on *Achlya glomerata*, upon which *O. Achlyae* was found to be obligate, but all were negative. Negative results were also obtained with the following water molds: *Sapralegnia ferax*, *S. delica*, *S. diclina*, *S. litoralis*, *S. megasperma*, *Aplanes treleaseanus*, *Dictyuchus missouri-*

ensis, *Isoachlya intermedia*, *I. itoana*, *I. unispora*, *Aphanomyces laevis*, *Protoachlya hypogyna*, *Achlya racemosa*, *A. americana*, *A. apiculata*, *A. colorata*, *A. caroliniana*, *A. crenulata*, *A. flagellata*, *A. recurva*, *A. proliferoides*, *Allomyces javanicus*, *A. moniliformis*, *Apodachlya pyrifera*. However, *Geolegnia inflata* became parasitized when put into a dish with the parasite.

Octomyxa Brevilegniae n. sp.

Obligate parasite in *Brevilegnia linearis* Coker and *Geolegnia inflata* Coker and Harvey, causing spherical galls mostly on the ends of the hyphae of the host. Mature galls 30–90 μ thick. Plasmodia of the parasite at maturity completely filling the galls and segmenting into zoosporangia; plasmodia developed later segmenting into resting spores in clusters of eight. Zoosporangia globose to ovoid, 5.2–7.6 μ , mostly 7 μ , in diameter, thin-walled. Spores discharged through papillae which are formed only on some of the sporangia next to the host wall, the other peripheral sporangia and those deeper within the sorus discharging their spores through those furnished with papillae; zoospores biflagellate with one long posterior and one short anterior flagellum, 4–6 in a zoosporangium. Resting spores 2.8–4.9 μ , mostly 3.5 μ , in diameter, with smooth, somewhat thickened walls; germination unknown.

Walls of the zoosporangia and resting spores show a negative cellulose reaction with chlor-iodide of zinc.

The Latin diagnosis follows.

Fungus parasiticus in hyphis *Brevilegniae linearis* et *Geolegniae inflatae*. Maturis tumoribus 30–90 μ in diametro. Plasmodiis plantae parasiticae maturitate tumores omnino complentibus. Primis plasmodiis formantibus zoosporangia; plasmodiis postea factis formantibus sporas perdurantes in massis octonas cuque massae. Zoosporangiis globosis vel ovoideis, parietibus tenuibus, 5.2–7.6 μ in diametro, plerumque 7 μ ; zoosporis biflagellatis cum uno longo flagello posteriore et uno brevi anteriore; 4–6 in zoosporangio. Sporis perdurantibus 2.8–4.9 μ in diametro, plerumque 3.5 μ , cum parietibus levibus leviter incrassatis; germinatione incompta.

SUMMARY

A new species of *Octomyxa* parasitic in *Brevilegnia linearis* and *Geolegnia inflata* is described.

Nuclear divisions of the developing plasmodium are of the so-called "proto-mitotic" type.

Zoospores are biflagellate with one long and one short flagellum.

The host range is limited, so far as known, to the above two species of water molds.

The writer is sincerely grateful to Dr. J. N. Couch for his encouragement and guidance and to A. W. Ziegler for water mold cultures.

LITERATURE CITED

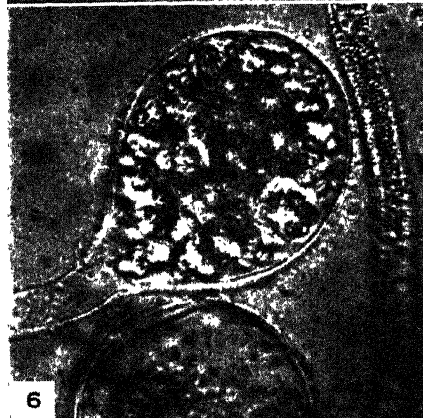
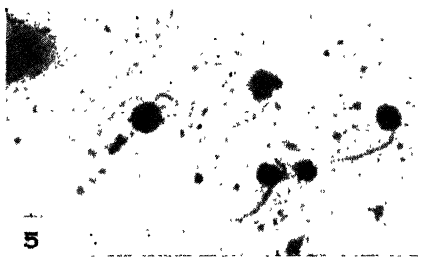
- COKER, W. C. 1927 Other Water Molds from the Soil. Jour. Elisha Mitchell Sci. Soc. 42: 207–226.

- COUCH, J. N. 1932 The Development of the Sexual Organs in *Leptolegnia caudata*. Amer. Jour. Bot. **19**: 584-599.
- , LEITNER, JEAN, AND WHIFFEN, ALMA. 1939 A New Genus of the Plasmodiophoraceae. Jour. Elisha Mitchell Sci. Soc. **55**: 399-408.
- 1941 The Structure and Action of the Cilia in Some Aquatic Phycomycetes. Amer. Jour. Bot. **28**: 704-713.
- HARVEY, J. A. 1925 A Study of the water molds and Pythiums occurring in the Soils of Chapel Hill. Jour. Elisha Mitchell Sci. Soc. **41**: 151-164.
- JOHANSEN, D. A. 1940 Plant Microtechnique. McGraw-Hill Book Co., New York.
- LEDINGHAM, G. A. 1934 Zoospore Ciliation in the Plasmodiophorales. Nature **133**: 534.
- 1939 Studies on *Polymyxa graminis* n. gen. n. sp., a Plasmodiophoraceous Root Parasite of Wheat. Canadian Jour. Res. **17**: 38-51.

EXPLANATION OF PLATE 15

- Fig. 1. Habit Sketch of *Octomyxa Brevilegniae* on *Brevilegnia linearis*. Parasite causing hypertrophy of the tips of the host hyphae. $\times 120$.
- Fig. 2. Immature plasmodium causing hypertrophy of hyphal tip. Host hypha five days old, plasmodium two days old. $\times 750$.
- Fig. 3. Plasmodium stained with Couch's modification of Gram's gentian violet technique. Vegetative nuclear divisions of the "protomitotic" type clearly visible in upper part of plasmodium. $\times 1080$.
- Fig. 4. Parts of three zoosporangial galls. Zoospores already formed within the zoosporangia in the gall at lower right. $\times 750$.
- Fig. 5. Biflagellate heterokont zoospores stained with the Löffler flagella-staining technique. $\times 1080$.
- Fig. 6. Host hypha filled with the octo- groups of resting spores. $\times 750$.
- Fig. 7. Groups of eight resting spores of the parasite free of the host hypha. $\times 750$.

PLATE 15



NOTES ON SOME HIGHER FUNGI

BY W. C. COKER

Department of Botany, University of North Carolina
Chapel Hill, North Carolina

PLATES 16-25

Hygrophorus Peckianus Howe. Bull. Torr. Bot. Club **5**: 43. 1874.

Camarophyllus Peckianus (Howe) Murrill. N. Amer. Fl. **9**: 389. 1916.

Plates 16 (inset) and 25, fig. 1

Cap 1-1.3 cm. broad, convex, not striate, dull, surface like leather or felt, not viscid, hygrophanous, light grayish buff when dry, deep smoky brown (blackish in places) when wet. Flesh 1 mm. thick, color of cap, very brittle, taste mild, odor keen and strong, remarkable and unlike that of any other mushroom, more like bad breath than anything else.

Gills thick, fleshy, color of unsoaked cap, distant, about 2 mm. broad, strongly decurrent by a broad tooth (arcuate).

Stem 2.5-3 cm. long, 1.6 mm. thick above, tapering downward, blackish below, color of gills above, smooth, granular at very top, stuffed, becoming hollow, pliable.

Spores (of No. 2620) subglobose to short-ellipsoid with a small excentric mucro, smooth, 3.7-4 x 4-5.5 μ .

This is a peculiar and rare plant, not previously reported south of New York and Michigan. It is easily recognized by its color, small size, and remarkable odor. The spores are not rough as described by Howe, Peck, and Murrill, the granular contents having a misleading effect. When the contents are plasmolyzed by salt solution the wall is seen to be perfectly smooth, as described by Smith and Hesler (Lloydia **5**: 12, 1942). *Hygrophorus Peckii*, which we also have in Chapel Hill, has a strong but very different musky odor and is strongly glutinous, etc. *Hygrophorus Peckianus* should be carefully compared with the English plant *H. foetans* Phillips (Grevillea **7**: 74, pl. 121, fig. B, 1878).

North Carolina. Chapel Hill. No. 2620. In rocky soil by path, mixed woods, July 9, 1917.

No. 2798. In mossy grass in a lawn, July 28, 1917. No. 5334. In damp soil in deciduous woods, July 10, 1922. Spores ovoid, 3.6-4.2 x 4.8-6.5 μ .

Hygrophorus amygdalinus Peck. Bull. Torr. Bot. Club **25**: 322. 1898.

Plates 16 and 25, fig. 2

Cap up to 3 cm. wide, when young nearly hemispheric, more or less gibbous with margin strongly inturned, at maturity plane in center with broadly drooping margin, surface gray-drab at all ages, viscid, fibrous, margin felted-spongy like the stem, representing an ephemeral veil which is an extension of the stem covering. Flesh pure white, firm, about 5-6 mm. thick near stem, rapidly thinning toward the margin; odor strong of bitter almonds; taste mild.

Gills pure white, about 2.5 mm. wide, rounded distally, pointed and either squarely adnate or more commonly slightly decurrent, rather distant and thick, about every other one short.

Stem up to 5 cm. long and 5 mm. thick in center, concolorous and with same surface as the margin of the young cap, minutely scurfy-squamulose, crooked, terete or somewhat grooved or flattened, stuffed with fibers which may separate, forming irregular hollows; base prevailingly pointed and yellowish but not always pointed and sometimes whitish.

Spores smooth, subellipsoid, proximal end somewhat narrowed, $5-6.2 \times 10-13\mu$ with one large droplet except when quite fresh. Basidia clavate, $7.5-9.5\mu$ thick, projecting up to 11μ , not counting the sterigmata which may be 11μ long, 4-spored (a few apparently only 2-spored). No true cystidia but some pointed sterile threads present.

Murrill (N.A.F. 9: 394) listed this species as a synonym of *H. hypothejus*, but we find it easily distinct in the characteristic odor, felted rather than glutinous cap margin and stem, pure white gills and larger spores. Dr. A. H. Smith has kindly confirmed our identification of this species.

We find no record of its occurrence except at the type station in the District of Columbia. Smith and Hesler have not found the species but have seen the types and make the following note (Lloydia 2: 58, 1939): "Sections of the type show that it is a *Limacium* and that its spores measure $9-12 \times 5-6.5\mu$. It needs further study."

North Carolina. Chapel Hill. No. 10698. In poor sandy soil on a low bank in pine woods, Nov. 14, 1937. No. 10700. Same spot as above, Nov. 21, 1937.

Hygrophorus rubropunctus Peck. Bull. N. Y. St. Mus. 116: 49. 1907.

H. glutinosus Peck. Bull. N. Y. St. Mus. 54: 950. 1902.

Plates 17 (bottom), 18, and 25, fig. 3

Cap up to 10 cm. broad, usually 5-7 cm., nearly pure white when quite fresh, becoming more or less buffy yellow, covered with thick gluten, as on lower part of stem.

Gills subdistant, adnate, decurrent by a line, nearly white, interveined in larger plants.

Stem short, stout, equal or tapering slightly upward, white, solid, not viscid above but lightly punctate there with concolorous dots which turn reddish on drying, glutinous below and strongly *scabrous-ridged*, base blunt.

Spores (of No. 13210) pure white, large, oval or subellipsoid, smooth, $5.6-7 \times 9.3-12\mu$. Basidia (of No. 3771) about 10.5μ thick, 4-spored.

Gregarious in small colonies. In unexpanded buttons the thick gluten can be seen to be creamy yellow.

Easily distinguished from *H. Laurae* by stout, blunt stem which is much roughened downward, by the red dots on upper stem when dry, and by the larger spores. We find no previous illustration.

North Carolina. Chapel Hill. No. 919. In thick woods under oak, Oct. 15, 1913. No. 932. Dry woods, Battle Park, Oct. 16, 1913. No. 3771. Deciduous woods by Battle branch and in barren clay spot in pines, Nov. 18, 1919. Spores oval, $5.2-$

7.4 x 9-11.2 μ . No. 4933. In woods, Dec. 7, 1921. Spores 5-6.7 x 7.4-11 μ . No. 13210. Ten plants; deciduous woods, Oct. 25, 1942.

Hygrophorus borealis Peck. Rept. N. Y. St. Mus. **26**: 64, 1874; Bull. **116**: 55, 1907.

Plates 17 (top) and 25, fig. 4

Gregarious and sparingly cespitose; cap (1) 2.5-3.7 cm. wide, strongly convex with drooping margin to irregularly plane, even or slightly depressed in center, hygrophanous, nearly pure white, watery white when moist, center at times faintly buff; margin even or substrate, neither cap nor stem at all viscid. Flesh a mere membrane except near center, white, not brittle, elastic; odor none, taste mild.

Gills distant, none forked, many short, interveined, arcuate-decurrent, about 4 mm. wide, thin, margin even.

Stem slender, crooked, irregular, usually collapsed here and there or down one side, about 5-7 cm. long and 6-8 mm. thick, subequal, base blunt and rounded or more or less pointed, surface quite smooth, fibrous, silky-shining, easily splitting, quite hollow or loosely fibrous-stuffed.

Spores (of No. 13209) pure white, oval-subpipshaped, with a prominent mucro, smooth, 5.2-6.5 x 7.4-10 μ . On standing in the herbarium the spores become a pretty clear yellow, about Pinard yellow (Ridg.). See Smith and Hesler (cited below) for variations in size and number of spores on a basidium.

This seems to agree perfectly with *H. borealis* Peck (*Camarophyllus borealis* in Murrill), and we have recently had our determination confirmed by Dr A. H. Smith. Murrill reports it from no farther south than New Jersey but it has since been reported from Tennessee by Hesler (Journ. Tenn. Acad. Sci. **20**: 235, 1945) and from North Carolina by Smith and Hesler (Lloydia **5**: 19, 1942). We have examined a plant from Fort Lee, New Jersey, in the New York Botanical Garden Herbarium (Earle, Sept. 15, 1902) as *H. borealis*, and find the spores exactly as in our No. 13209 and also as in Hesler's collection from Tennessee.

We cannot find that the species has previously been illustrated.

North Carolina. Chapel Hill. No. 13209. In deciduous woods about 7 miles west of town, Oct. 25, 1942. Also Nos. 1974, 3756, 3762, 3778, 3870, 3890 from Chapel Hill.

Tennessee. Cades Cove. Hesler, No. 16574. Spores 5-6(6.8) x 7-9.3 μ .

New Jersey. Fort Lee. Ellis, coll. (N. Y. B. G. Herb.).

H. borealis forma **salmonaeus** n.f.

Plate 25, fig. 5

Cap flushed with pinkish orange (light salmon) on one side and stem dull pink below, the pink spreading when plants are handled. Spores 4.3-5.5 x 7-10 μ .

No. 3756 (entered above) was faintly pinkish on a part of the margin and stem turned faintly pink when rubbed.

North Carolina. Chapel Hill. No. 3745. Mixed woods, Nov. 13, 1919.

Craterellus calyculus (B. & C.) Burt. Ann. Mo. Bot. Gard. 1: 338. 1914.

Stereum calyculus B. & C. Hooker's Journ. Bot. and Kew Misc. 1: 238, 1849; Grevillea 1: 161. 1873.

Thelephora subundulata Peck. Bull. Torr. Bot. Club 22: 492. 1895.

Craterellus subundulatus Peck. Bull. N. Y. St. Mus. 67: 27. 1903.

Craterellus albomarginatus Coker. Journ. Elisha Mitch. Sci. Soc. 43: 139. 1927.

Plates 19 (bottom, right) and 25, fig. 6

In the Mitchell Journal above cited we published under the name *Craterellus albomarginatus* a little plant from Linville Falls, North Carolina. Later, in looking through the genus *Craterellus* at the New York Botanical Garden, we found a collection by Peck and Earle of *C. subundulatus* and recognized it as the same species. It was not included in Burt's monograph but was later recognized in his supplement (Ann. Mo. Bot. Gard. 13: 312. 1926). We have also studied an ample collection of Chapel Hill plants and have come to the conclusion that they all belong to the species *C. calyculus*. The plants vary considerably in width of cap and irregularity of margin, and the hymenium dries more yellowish in some cases.

The species was originally described from South Carolina, and has been reported from North Carolina and (as *C. subundulatus*) from Delaware and New York, and Tennessee. In the Coweeta Experiment Forest in Macon County, N. C., we found a specimen that had assumed a petaloid shape from a nearly marginal stem, due apparently to its growth from the side of a bank.

We give below a slightly revised description.

Cap 4-15 mm. wide, deeply depressed to infundibuliform, cream buff to a darker buffy or grayish brown, dull, dry, inherently fibrous and radially roughened, usually with hispid squamules on marginal half, which is often bent downward and strongly crumpled. Flesh delicate, pliable, with a faint taste and odor like that of *C. cibarius*.

Hymenium pale buff to cinereous buff, tender, slightly rugulose, uneven, decurrent.

Stem about 1-2 cm. long and 1-1.5 mm. thick, smooth, glabrous, firm, pliable, concolorous with cap and with a small hollow; base not noticeably tomentose.

Spores (of No. 7464) internally granular when fresh, smooth, ellipsoid, 4.8-6 x 6.5-8.5 μ ; basidia long-clavate, most 4-spored but often 6-spored, 6-7.4 μ thick and up to 35 μ long.

In drying the cap color tends to become more cinereous and the hymenium ochraceous, and stems that collapse before drying are black. The entire plant is strongly hygroscopic, and after drying will regain its full size and shape in less than a minute. This character of reviving is a key character used by Murrill (N. Amer. Fl. 9: 163) in separating *Plicatura* and *Xerotinus* from other genera of the tribe. He does not include this species, which was since recognized by Burt as a *Craterellus*.

Our plants are much smaller than *C. sinuosus* as shown by Fries (Icones 2: pl. 196, fig. 2) and both cap and hymenium are more yellow; also the spore meas-

urements are distinctly smaller than those of *C. sinuosus* as given by Burt (6-7 x 10-12 μ).

North Carolina. Chapel Hill. No. 7464. On damp clay soil, July 31, 1924.

Linville Falls. No. 8206. Type of *C. albomarginatus*. On bare earth just above the water of a spring, hemlock-rhododendron woods, Sept. 10, 1927.

Macon County. No. 12154. On bank by trail in Coweeta Experimental Forest, Aug. 12, 1939.

Psalliota pocillator Murrill. *Mycologia* 33: 446. 1941.

Plates 20, 21, and 25, figs. 8-10

Cap up to 8.5 cm. broad, subovate, soon broadly conical and gibbous, expanding to plane with a gibbous center, the margin involute; color when young brown all over or the margin paler and center darker, soon dark sepia on the broad umbo, rather abruptly fading to light buffy brown (about pale avellaneous of Ridg.); surface glabrous and smooth, subshining. Flesh nearly white, delicate, not brittle, very thin, about one mm. thick nearly to center where it abruptly thickens to 5 mm., a mere membrane at the margin, turning slowly dull red when bruised (not when cleanly cut); odor peculiar, like stale meat or fish, taste unpleasant, causing flow of saliva and leaving a sticky feel in the mouth.

Gills free, close, narrow, about 5 mm., delicate, margin even, whitish, soon pale pink then darker to about color of cap center.

Stem long and slender, nearly equal, more or less crooked, 3-7 mm. thick and up to 13 cm. long, very smooth and shining above and below the veil, inserted for a short distance into the humus and soil and ending abruptly in a compressed bulb which is concave above and surrounded with a thick, blunt, raised margin like the saucer for a flower pot; stem white at first then more or less pale avellaneous except in the soil where it remains white; bulb connected with one or several slender white strands; stem flesh firm and rigid and very solid except for a small, ill-defined, loosely fibrous central column. Veil ample and permanent, pale then brown, attached at about 8-10 mm. from the top, thin but set below with spongy flecks and often divided into more or less complete layers.

Spores (of No. 13587) dark sepia (no trace of purple), oval, 2.8-3.5 x 4-5.5 μ . Basidia clavate, 4-spored, about 6.7 x 22 μ ; cystidia none.

This is a peculiar plant. In form and stature it is much like *P. abruptibulba* but differs strikingly from it in the abruptly dark umbo, the entirely glabrous stem, in change to reddish, not yellow, when bruised, the unpleasant taste, and the very small spores, and from that and all other species in the cup-shaped bulb. At no age is there any connection between the rim of the bulb and cap or stem, the very young button sitting in the bulb like a ball in a cup. The odd shape of the bulb is more obvious where the plant is growing in light leafy humus. When in earth it is obscured by the attached soil. The colors of the cap are well retained in drying, the abruptly dark brown center contrasting strongly with the much lighter color of the surrounding surface. In the first collection there were 40-odd plants in cespitose groups or single over an area of several square yards.

This striking plant was described from Gainesville, Fla., and so far as we know has not previously been reported from anywhere else. It is strange that so conspicuous and prolific a species should have been so rarely found. Murrill says

"excellent for the table," but our well known mycophagist, Prof. N. B. Adams of Chapel Hill, finds it inferior to others of the genus.

North Carolina. Chapel Hill. No. 13587. In humus under shrubs in the Arboretum, July 21, 1944. No. 13593. Same place in Arboretum, Aug. 11, 1944. No. 13602. Same place as above, Sept. 25, 1944. About 30 plants, all stages, in a compact group, some fused at base. No. 13608. In scattered clusters from same colony as above, Sept. 29, 1944. These plants were water-soaked from rains and showed considerable rose tint in the cap.

Hydnellum diabolus Banker. *Mycologia* 5: 194. 1913.

Hydnellum carbunculus (Secr.) Banker. Mem. Torr. Bot. Club 12: 151.

1906. Not *Hydnum carbunculus* Secr. *Myc. Suis.* 2: 151. 1833.

Cap 5-11.5 (15) cm. broad, convex, uneven, surface of growing parts soft, taking the imprint of a finger, white, turning red-brown when rubbed or exposed, then through darker brown to almost black with some whitish or pale fawn areas left here and there, especially on the margin; fresh, damp, growing parts secreting small droplets of clear bright red juice which disappear, leaving dark red-brown stains. Flesh of cap thick, soft throughout or somewhat harder downward and strongly zoned with dark and light brown layers when fresh, far less conspicuously so when dry; taste quite acrid, odor marked but not strong, medicinal or of old hickory nuts, not unpleasant.

Spines very short, 3 mm. long or less, crowded, light fawn color with white tips, then dark brown, not decurrent.

Stem short, 2-4 cm. long, 1-1.5 cm. thick, tough, flesh dark brown to blackish, with no distinct spongy layer but outer flesh softer.

Spores (of No. 14081) light fawn color, between avellaneous and vinaceous buff (Ridg.), subglobose to oblong in general outline, distinctly and rather irregularly warted, 3.7-4.2 x 4.2-5.4 μ .

Our plants, which are usually found in pine straw, grow around the needles and imbed them within the fungus body. The plants shrink very little in drying. A bit of the flesh put in water turns it reddish.

Illustrations: Coker. *Journ. E. M. Sci. Soc.* 34: pls. 14, 15, 28. 1919.

North Carolina. Chapel Hill. Nos. 1341, 1869, 1951, 1968 (as published in *Journ. Mitch.*

• Soc. cited above). No. 3811. Pine woods, Dec. 2, 1919. No. 14056. In pine straw, east bank of University Lake, Oct. 16, 1946.

Chatham County. No. 14081. Growing up through dense pine straw under *Pinus taeda* in sandy old field, Mann's Chapel, Nov. 4, 1946. No. 14299 and No. 14301.

In pine grove, near same place as above, Oct. 18, 1947. No. 14311. In second growth pine grove by road to Mt. Carmel, Oct. 19, 1947.

Durham County. No. 14326. Under pines near New Hope Creek, Oct. 23, 1947.

Hydnellum suaveolens (Scop.) Karst. -*Medd. Soc. Fauna et Fl. Fenn.* 5: 27. 1879.

Hydnum suaveolens Scop. *Fl. Carn.* 2: 472. 1772.

Hydnum compactum Pers. *Comm. Schaeff.*, p. 57. 1800.

Hydnum boreale Banker in White. *Bull. Torr. Bot. Club* 29: 553. 1902.

Sarcodon gravis Coker. *Journ. Elisha Mitch. Sci. Soc.* 55: 376. 1939.

Plate 19 (top, and bottom, left)

Plants single or in colonies, at times confluent; cap (3.6) 6–12 (14) cm. wide, circular, the thick sterile margin more or less uneven; surface convex then plane, more or less scrobiculate, at times strongly so, pure white all over at first and remaining so toward the margin until old, very soft-felted, taking the imprint of a finger, the felt collapsing in center and gradually outward to a soft, smooth, tan-brown surface which may be faintly tinted with violet, fawn, or olive. Flesh soft above, gradually harder downward, conspicuously zoned with soaked drab and white layers, the drab becoming blue or greenish, especially downward, on exposure. Taste none; odor variable in intensity, faint when first collected but after a few hours usually distinctly aromatic, as of wintergreen or peppermint, lost for the most part in drying.

Stem short, about 1–2 cm. long and up to 1.5 cm. thick, surface firm, without a superficial mass of tomentum, deep indigo blue or blackish or blue-black; mycelium blue, at least at times; flesh solid, strongly and closely zoned with deep blue, even when freshly cut.

Spines crowded, up to 6 mm. long, pale drab, then drab-brown with pale tips, often descending far down the stem, at times even to the substratum.

Spores (of No. 13324, good print) fawn-colored, irregularly angled and bluntly warted, 3–3.7 (4.2) \times 3.8–5 μ . Basidia clavate, 4-spored, about 4.8–5.2 μ thick.

Sarcodon gravis, as described by me from Highlands, N. C. (see above), is this species. Since that time we have found a number of plants in several consecutive years, evidently from the same colony. They are all essentially alike. Our plants show less vivid blue than some of the European and northern examples, but the blue color is quite evident and all other characters are the same.

This species seems not to have been found heretofore in the east except for Canada and Maine. For a western form, see entry under Wyoming.

Illustrations: Coker. Journ. E. M. Sci. Soc. 55: pl. 35 (top) and pl. 44, fig. 4 (as *S. gravis*). 1939.

North Carolina. Highlands. No. 10563. By road around Ravenel Lake, Aug. 19, 1937 (type of *Sarcodon gravis* Coker). No. 11740. Beside stream near entrance to Rhododendron Trail from road by Ravenel Lake, July 20, 1940. No. 12822. Same spot as above, under hemlock and rhododendron, July 12, 1942. No. 12834, same spot (remainder of large colony), July 15, 1942. No. 12874. Same spot as above, July 27, 1942. No. 13317. Same place as above, July 24, 1943. No. 13324. Same place as above, July 26, 1943. No. 13487. Under rhododendron and hemlock, just across road from entrance to Rhododendron Trail, Aug. 17, 1943.

Maine. Mt. Desert Island. Dr. Fred J. Seaver, coll. (N. Y. B. G. Herb. and U. N. C. Herb.)

Wyoming. Albany County. No. 14699. On duff in coniferous forest, altitude 9500 feet, Medicine Bow Mts., Sept. 6, 1940. Ragnhild and W. G. Solheim, coll. (Univ. of Wyo. and U. N. C. Herb.) Spores irregularly angled and warted, 4–5.2 \times 4.2–6.5 μ . These plants differ in larger spores, and stouter, heavier form of the plant itself. It may be the same form as reported and illustrated by Kauffman from Tolland, Colorado (Pap. Mich. Acad. Sci. 1: 116, pl. 33, 1921).

Canada. Quebec. Under balsam, St. Aubert, July 15, 1940. H. A. C. Jackson, coll. (No. 1832 in Snell Herb.)

Sistotrema confuens Pers. Disp. Meth. Fung., p. 28, 1797; Syn. Meth. Fung., p. 551. 1801.

Hydnum sublamellosum Bull. Herb. Fr., pl. 453, fig. 1. 1789.

Plate 22 (top)

We first published notes on this species with illustrations in the Journal of the Elisha Mitchell Scientific Society **41**: 285, pls. 63, 65, figs. 8-11, 1926, having found it twice in Chapel Hill on pine needles. Since then we have found it abundantly in several colonies in fine condition. Our photograph shows a thick mat of needles (*P. echinata*) with the fungus growing up among them. The perfectly fresh unweathered plants are at first finely tomentose with slender hairs, soon practically glabrous and slightly viscid.

Some European illustrations, particularly Bulliard's and Greville's, show a plant in size and appearance hardly like our delicate form, but others, as W. G. Smith's fig. 87, are more like ours. Patouillard's figure (Hymen. Eur., pl. 3, fig. 19) shows the 6-spored basidia. Konrad and Maublanc (Icon. Sel. Fung. **5**: pl. 426, fig. 1) show the species with bright red stalks and place it, as does Patouillard, in the Polyporaceae.

We have specimens in our herbarium as follows: North Carolina (Chapel Hill), Tennessee, Virginia, New Hampshire, and Ontario. It has also been reported from North Carolina by Schweinitz, Alabama (Earle, Banker), New York (Peck), and Vermont (Burt). Our photograph was made from the collection listed below.

North Carolina. Chapel Hill. No. 13241. Second growth pine grove, Nov. 7, 1942. Seven or eight colonies were seen, most of them forming segments of rings; one perfect ring about 3½ ft. in diameter. The appearance at a little distance was as if crumbled chalk had been dropped in the pine needles.

Hydnodon thelephorum (Lév.) Banker. Mycologia **5**: 297. 1913.

Plates 22 (bottom) and 25, fig. 11

Since our first report of this peculiar tropical plant in Chapel Hill (Journ. E. M. Sci. Soc. **41**: 279, pls. 56 and 64, figs. 16, 17. 1926), we have found other fine examples of it, always near the bases of deciduous trees on banks by branches. Our plate shows a part of two of these collections. In a later collection (No. 13835) one of the fused clusters reached a size of 5 x 6.5 cm. Good prints show the spores to be coral pink (Ridgway). A few thin-walled cells with swollen bases were found among the basidia (fig. 11). Basidia, spores, etc., as described before.

North Carolina. Chapel Hill. No. 13809 and 13810, Aug. 17, 1945. No. 13835, Aug. 24, 1945 (W. C. C., coll.).

Boletus Ananas M. A. Curtis. Amer. Jour. Sci., ser. 2, **6**: 351. 1848.

Plate 23 (top)

Cap strongly convex or center flat, up to 7.5 cm. broad (not counting veil), covered all over with a thick shaggy thatch which is a fine terra cotta pink below, the thick outer fibers paler, almost white, this thatch expanding at the margin.

as a tough veil membrane which is pink below, the paler fibers extending as torn flaps for as much as one to two centimeters. Flesh of cap up to 1.5 cm. thick, firm, durable, not grubby, white, instantly blue when cut, soon white again, then dull pinkish leather color on standing; taste mildly acid, odor none.

Tubes mustard yellow, turning to deep reddish brown, then dark blackish brown, blackish on bruises, dark green-blue when cut, then brown; up to 1.5 cm. deep, mouths even, about 1 mm. wide, strongly depressed at stem.

Stem, with bulb, up to 11 cm. long, subequal, 12-14 mm. thick in center, smooth, white, the very tip with a sharp bright pink ring about 5-7 mm. wide, then brownish when exposed.

Spores (of No. 13756) in light print brown with faint purplish tint, in heavy print blackish brown, strongly striate, ellipsoid, 6-7 x 13.5-15.5 μ .

Bulb about 2-2.5 cm. thick, extending as plates or fibers apparently connected to rotten wood. In No. 13756 one plant was from the base of an oak and one among roots of pine.

Illustrations: Coker and Beers. Boletaceae of North Carolina, pl. 64, fig. 11.

South Carolina. Myrtle Beach. No. 13756. July 13, 1945. No. 13779. Attached to bark at base of a pine tree, July 26, 1946. Spores dark brown, ellipsoid, strongly striate, 6.3-7.5 x 13.6-17.3 μ .

Boletus sphaerosporus Peck. Bull. Torr. Bot. Club 12: 33. 1885.

Plates 23 (bottom) and 25, fig. 12

Five large heavy plants about 8-13 cm. broad, plano-convex, margin strongly interned and distinctly free, for as much as 5 mm. in places; surface quite glabrous, shining, with a viscid removable cuticle, not acid to the tongue, color reddish brown, near russet (Ridg.), becoming much darker, blackish brown when rubbed. Flesh thick, up to 3.5 cm. at stem, thinning rapidly near margin and extending as a thick free membrane, white, then quickly brown when exposed, tender but firm; taste slight, not pleasant, somewhat flat and alkaline, odor negligible.

Tubes up to one cm. long, easily removable, large, very variable in size, 2-3 mm. in diameter toward margin, mouths much elongated radially near stem where they may be 8 mm., inset with smaller, more shallow ones, depressed at stem and descending for several mm. as reticulations or scarcely depressed in places.

Stem stout, more or less plug-like, rounded below into a blunt base, length up to 6.5 cm. and thickness 3.4 cm. near top, glabrous, color dull pale yellow above, concolorous with cap below. Flesh light brown, then darker brown when cut.

Veil very thick and conspicuous, remaining attached about the middle of the stem as a heavy roll collapsed upward, pallid whitish when fresh, becoming discolored to dark brown.

Spores pale yellow-brown under microscope, subglobose, smooth, 6-7.4 x 7-9 μ , with a distinct oil globule.

This peculiar and very interesting plant was described by Peck from one dried plant from Wisconsin (Bull. T. B. C. 12: 33. 1885). He discusses its peculiarities at some length and is particularly concerned with the prominent veil. In our plants this is obviously a veil and not a volva. This heavy veil and the unique subglobose spores led him to suggest that it might be a sufficient distinc-

tion to consider it a new genus. Murrill (N. Amer. Flora **9**: 155) reports it from Wisconsin, Iowa and Minnesota.

Illustrations: Peek. Bull. Torr. Bot. Club **12**: pl. 49, figs. 1, 2. 1885.

Minnesota. St. Paul. T. Kommedahl. (Received by us from him by air mail Oct. 9, 1945. No. 13994.)

Boletus placidus Bon. Bot. Zeit. **19**: 204-205. 1861.

B. albus Pk. Rept. N. Y. St. Cab. **23**: 130. 1873.

Plate 24

Since the appearance of our book on the Boletaceae we have collected good material of this species at Highlands, N. C., and are giving below a more adequate description, with photographs. Three of these collections grew from the same mycelium in a damp place (rhododendrons, hemlocks, etc.) by a brook north of the laboratory. The photograph shows a peculiarity noted in all the plants from this spot. The pore surface, while perfectly functional, shows odd pits and cavities of different sizes and depths. These are almost certainly not caused by insects as the plants showed no signs of insects and the appearance is different from that of insect attacks on other boletes. Also it is hardly to be expected that if caused by insects this peculiarity would appear on all of the individual collections from this spot, made at different times.

Cap up to 9.5 cm. wide, convex then gibbous, glutinous, *pure white* when young, then lightly tinted with lemon yellow toward margin, quite smooth, no flecks or fibers, margin very narrowly free. Flesh thick in center, nearly white then lemon tinted toward tubes and margin; no taste or odor.

Tubes short, up to 5 mm., adnate, small, angular, walls very thin; color pale creamy yellow, then dull straw yellow; when young with droplets of nearly clear viscid fluid which turn light pink then brownish; tube surface always uneven, deeply and irregularly pitted, in some cases with pits so deep as to expose the cap flesh.

Stem 6-8.5 cm. long, 8-13 mm. thick, solid, nearly equal, more or less crooked, dotted all over with pale, then brownish pink droplets; base of stem nearly always bent and pointed, inserted into pure white, ropy mycelium.

Spores (of No. 13275) pinkish clay color in a good print, ellipsoid, smooth, 3-3.7 x 5.6-7.5 μ

This species is easily distinguished from all the other white or whitish boletes by the very dark, reddish brown color of the dried plants and by the spores.

North Carolina. Highlands. No. 8970. Under white pines, July 29, 1931. No. 11739.

In woods, mostly white pines, by Ravenel Lake, July 20, 1940. No. 12707. Under white pines, Aug. 26, 1941. No. 13275. A fine colony extending over 6 or more feet, single or two or three together, by a branch north of the laboratory, July 13, 1943. No. 13289. Same place as above, July 19, 1943. Spores 3-3.6 x 6.8-8(9), longer than in the earlier collection from the same place. No. 13579. Same spot as above, Sept. 6, 1943. Only one plant; cap nearly pure white but creamy in places. Spores 3-3.6 x 6-8 μ .

Cyphella cupulaeformis B. and Rav. *Grevillea* 2: 5. 1873.

Plate 25, figs. 14, 15

A very large collection, crowded by scores in places, was made from the bark of six living cedar trees in the Negro cemetery, Chapel Hill, N. C., Nov. 3, 1946 (No. 14087). Notes on this species have been published once before by us (*Mitchell Journal* 36: 150, pl. 30, fig. 3, 1921).

Cups attached by their sides and hanging vertically. Spores (good spore print on a slide) shaped like "Jack rocks," a few showing only two or three papillae but most with four in outline, making the spore "squarish" or slightly rectangular, $4-5 \times 5-6\mu$, not counting the projections, otherwise up to $6.5 \times 7.4-8\mu$. Threads of flesh $3.4-4\mu$ thick, with many clamp connections, anastomoses and some branching.

The species seems to have been reported only a very few times since it was collected by Ravenel in South Carolina and Georgia. Among the localities recorded are the Great Smoky Mountains National Park (*Mycologia* 33: 574, 1941) and Bermuda (*Mycologia* 38: 186, 1946).

Rhizopogon truncatus Linder. *Rhodora* 26: 196, pl. 148, fig. 3, 5-7. 1924.

Plate 25, figs. 16, 17

In August 1937 we found a very beautiful little *Rhizopogon* on an exposed road-side bank at Highlands, N. C. (No. 10511). There were two plants, about 1 cm. thick, bright sulphur yellow all over at first, then clear wax yellow when dry; peridium very thin; gleba light ashy color, with cavities so small as to be scarcely visible to the eye; odorless and tasteless. Spores ellipsoid, $3.5-4 \times 7-8.3\mu$, with a conspicuous cup. Basidia about 5μ thick and 13.5μ long.

In all essential characters this agrees with *R. truncatus*, which has been known before only from the type locality in New Hampshire. See Coker and Couch, *Gasteromycetes*, p. 36, pl. 26 (top) and pl. 106, fig. 13, 1928.

EXPLANATION OF PLATE 25

Fig. 1. *Hygrophorus Peckianus*. No. 2620. Spores.

Fig. 2. *H. amygdalinus*. No. 10700. Spores.

Fig. 3. *H. rubropunctus*. No. 13210. Spores.

Fig. 4. *H. borealis*. No. 13209. Spores.

Fig. 5. *H. borealis forma salmoncus* n.f. No. 3745. Spores.

Fig. 6. *Craterellus calyculus*. No. 7464. Spores.

Fig. 7. *C. calyculus*. No. 7464. Basidia, showing variation in number of sterigmata.

Fig. 8. *Psalliota pocillator*. No. 13602. Outline of one young plant, part of another, and (center) a very young button in its cup.

Fig. 9. *P. pocillator*. No. 13587. (Above) Base of stem with cup; (below) section through two fused bases, the inner margin of cup also fused.

Fig. 10. *P. pocillator*. No. 13587. Spores.

Fig. 11. *Hydnodon theleporum*. No. 13809. Hymenial cell with swollen base.

Fig. 12. *Boletus sphaerosporus*. No. 13994. Spores.

Fig. 13. *Cyphella cupulaeformis*. No. 14087. Spores.

Figs. 14, 15. *C. cupulaeformis*. No. 14087. Showing bit of hypha with clamp connection and a basidium with two sterile cells.

Figs. 16, 17. *Rhizopogon truncatus*. No. 10511. Basidium and spores.

Figures by Alma Holland Beers except figs. 8 and 9, which are by Alleda Burlage. All spore drawings $\times 1620$; figs. 7, 11, 14, 15, 16 $\times 810$; figs. 8, 9 $\times \frac{3}{4}$.

PLATE 16



Hygrophorus amygdalinus. No. 10700.
Hygrophorus Peckianus (inset). No. 2620. Nat. size.

PLATE 17



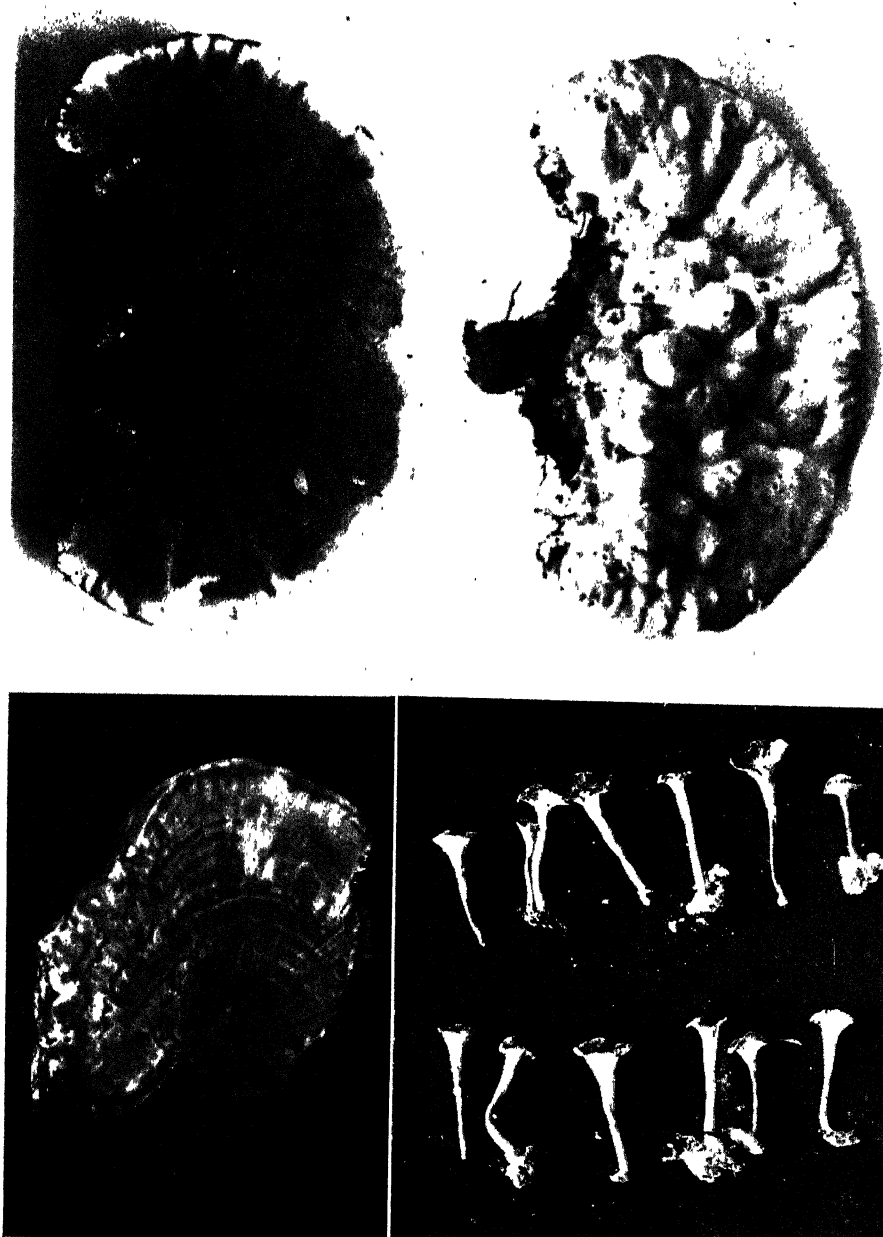
Hygrophorus borealis (top). No. 13209.
Hygrophorus rubropunctus (bottom). No. 13210.

PLATE 18



Hygrophorus rubropunctus: No. 3771.

PLATE 19



Hydnellum suaveolens. No. 13317 (top), $\times \frac{3}{4}$; No. 11740 (bottom, left), nat. size.
Craterellus calyculus (bottom, right). No. 7464.

PLATE 20



Psalliota pocillator. No. 13587. Photograph by J. N. Couch.

PLATE 21



Psalliota pocillator. No. 13587. Photograph by J. N. Couch.

PLATE 22



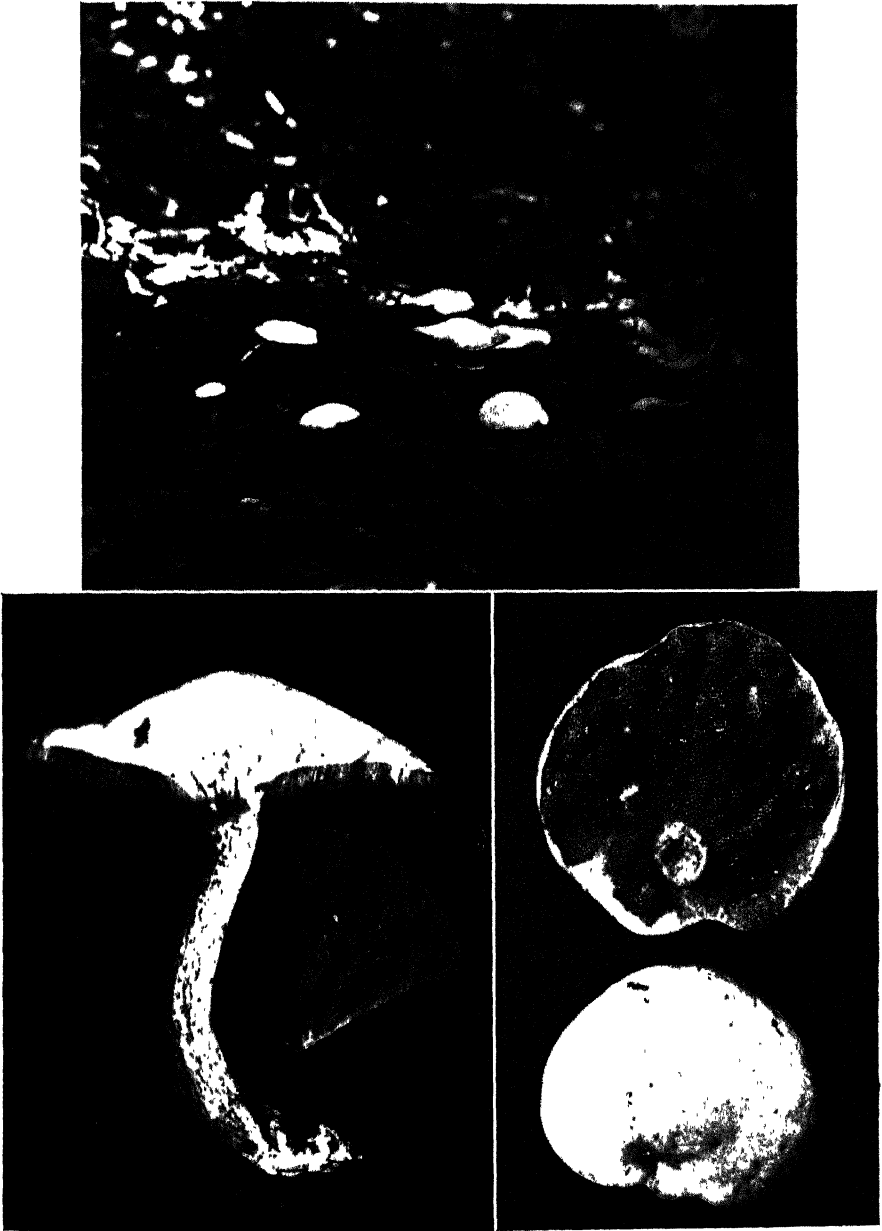
Sistotrema confluens (top). No. 13241.
Hydnodon theleporum (bottom). Nos. 13809 and 13810.

PLATE 23



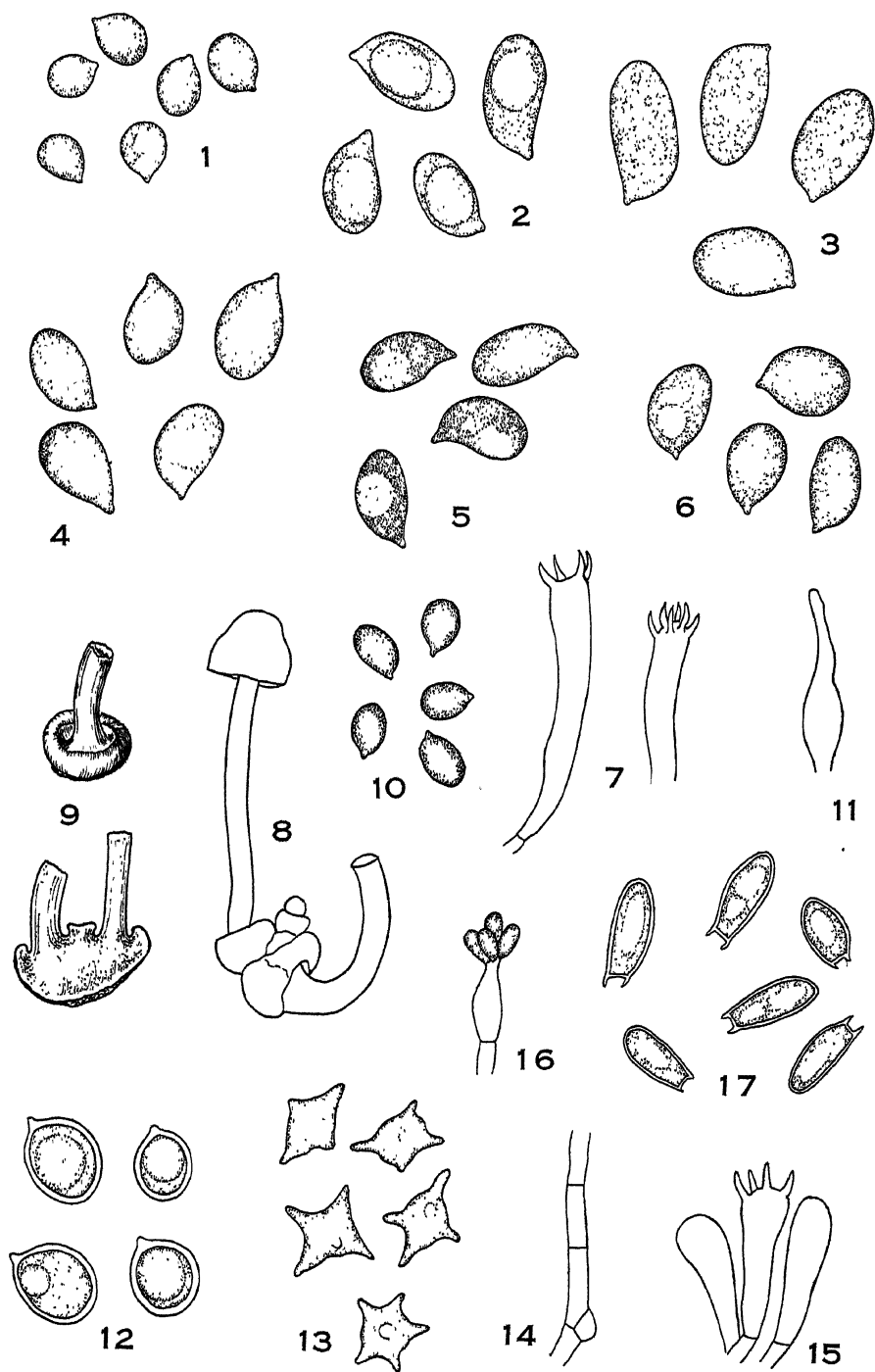
Boletus Ananas (top). No. 13756. Myrtle Beach, S. C. Photograph by Dr. Hugh Smith.
X about $\frac{1}{2}$.
Boletus sphaerosporus (bottom). No. 13994. St. Paul, Minnesota. Slightly reduced.

PLATE 24



Boletus placidus. Highlands, N. C. (Top) *in situ*; (bottom) No. 13289. Slightly reduced.

PLATE 25



JOURNAL
OF THE
Elisha Mitchell Scientific Society

Volume 64

December 1948

No. 2

PROCEEDINGS OF THE FORTY-FIFTH ANNUAL MEETING OF
THE NORTH CAROLINA ACADEMY OF SCIENCE

DAVIDSON COLLEGE, DAVIDSON, N. C.

The forty-fifth annual meeting of the North Carolina Academy of Science was held at Davidson College on May 7 and 8, 1948.

The Academy convened in a general session at 11:00 a.m. on the first day with President F. G. Hall presiding. After the presentation of five papers the session adjourned for lunch at 12:30.

The second general session was called to order at 2:00 p.m. the same day. After the reading of five papers the session closed and was followed by the business meeting.

The minutes of the 1947 meeting were approved as published in the Journal of the Elisha Mitchell Scientific Society **63**: 83-107. The reports of the various committees were then presented as follows:

REPORT OF THE EXECUTIVE COMMITTEE

The Executive Committee met in Durham, N. C., on May 6, 1948, and at Davidson College on May 7, 1948. At these meetings the following matters were transacted:

1. The Treasurer was authorized to increase the savings account to \$100.00 when feasible.
2. The Treasurer was authorized to spend up to \$100.00 for secretarial aid.
3. The Treasurer was authorized to pay all outstanding bills for operating expenses.
4. The Committee authorized the Treasurer to submit his financial report as of July 1, 1948, to the auditing committee.
5. The following were elected to membership in the Academy:

Ackerson, Robert Henry, Mathematics, Catawba College
Aderholdt, Daniel Franklin, Chemistry, Catawba College
Allison, Ray, Wildlife, N. C. State College
Banham, Katharine May, Psychology, Duke University
Barkalow, Frederick S., Wildlife, N. C. State College
Berryhill, Louise Russell, Geology, University of North Carolina

Beta Rho Chapter of Tri Beta, Wake Forest College
 Britt, H. Grady, Biology, Wake Forest College
 Carter, Donald Sanford, Chief Clinical Psychologist, State Hospital, Raleigh
 Chestnut, A. F., U. N. C. Institute of Fisheries, Morehead City
 Cobb, Whitfield, Mathematics, Guilford College
 Cornatzer, William Eugene, Biochemistry, Bowman Gray School of Medicine
 Critcher, Titus Stuart, Wildlife, N. C. State College
 Daggy, Tom, Biology, Davidson College
 Dixon, James Cannon, Psychology, University of North Carolina
 Dugger, Willie Mack, Jr., Botany, N. C. State College
 Fichter, George Siebert, University of North Carolina
 Godfrey, Robert Kenneth, Botany, N. C. State College
 Green, Paul Eliot, Jr., Agric. Engineering, Experiment Station, Oxford
 Haff, Mildred W., Mathematics, Elon College
 Haff, Richard Monroe, Natural Science, Elon College
 Hamnett, William Lawrence, Wildlife Resources Commission, Raleigh
 Harmon, Fannie Rosa, Biology, Flora Macdonald College
 Helms, Rufus Marshall, Physics, N. C. State College
 Howell, Charles DeWitt, Biology, Catawba College
 Hueske, Edward Ernest, Wildlife Resources Commission, Fayetteville
 Hunter, Wanda Sanborn, Zoology, Duke University
 McDowell, Gladstone Wadley, Hydraulic Engineer, Atlanta, Georgia
 Morgan, Mary Elta, Chemistry, Central High School, Charlotte
 Olds, Hayden Whitney, Wildlife Resources Commission, Raleigh
 Outten, Lora Milton, Biology, Mars Hill College
 Rhodes, Mary Brickert, Biology Librarian, Duke University
 Rhodes, Stanley Allen, Zoology, Duke University
 Roberts, Henry S., Jr., Zoology, Duke University
 Roelofs, Eugene Woodrow, U. N. C. Institute of Fisheries, Morehead City
 Rogers, Hollis J., Biology, Woman's College, U. N. C.
 Sommer, Roy Frederick, Physics and Chemistry, Pfeiffer College
 Stephen, William Archibald, Entomology, N. C. State College
 Van Dyke, Louise Amelia, Mathematics and Physics, Flora Macdonald College
 Ward, Margaret Clapman, Zoology, Duke University
 White, William A., Geology, University of North Carolina
 Wilbur, Karl Milton, Zoology, Duke University
 Williams, Annie John, Mathematics, Julian S. Carr Junior High School, Durham
 Wilson, Kenneth Alden, Wildlife Resources Commission, Currituck
 Woodhouse, Charles Bailey, Wildlife Resources Commission, Raleigh
 Wright, C. S., Phipps and Bird, Richmond, Virginia

6. The following were reinstated as members in the Academy:

Adams, Louise, Mathematics, High Point College
 Cell, John W., Mathematics, N. C. State College
 Ellington, Mary Oliver, Biology, St. Mary's School and Junior College, Raleigh
 Hill, Michael Arendell, Jr., Mathematics, University of North Carolina
 McCavock, W. G., Mathematics, Davidson College
 Mebane, William Nelson, Jr., Mathematics, Davidson College
 Schaeffer, Florence Louise, Chemistry, Woman's College, U. N. C.
 Straley, H. W. III., Geologist, Washington, D. C.
 Swanson, Marjorie Angela, Biochemistry, Bowman Gray School of Medicine
 Whitehead, Laurence C., Fish and Wildlife Service, Raleigh
 Williams, Maude, Biology, Woman's College, U. N. C.
 Wood, William Woodhull, Mathematics, Davidson College

7. The Secretary reported the following losses of members:

1. By resignation: 3
2. By non-payment of dues: 30
3. By death the following:
 - John Bewley Derieux
 - Franklin Sherman, Jr.
 - Charles Burgess Williams

8. The Committee accepted the invitation of the University of North Carolina to meet there in 1949.

The above reports of the Executive Committee were adopted by the Academy.

TREASURER'S REPORT

Financial Statement as of July 1, 1948

Receipts

Bank Balance (from July 1, 1947).....	\$45.42
Gifts	
Ornithological Essay Prize, 1948.....	20.00
N. C. Section American Chemical Society, 1947.....	5.00
A. A. A. S. Research Grant, 1947.....	128.50
A. A. A. S. Research Grant, 1948.....	65.00
Dues, 1947.....	18.00
Dues, 1948.....	474.50
Initiation fees, 1948.....	116.00
Total.....	872.42

Expenditures

Elisha Mitchell Scientific Society.....	300.00
Secretarial aid.....	87.50
E. C. Cocke (1947 A. A. A. S. Research Grant).....	64.25
T. L. Quay (1947 A. A. A. S. Research Grant).....	64.25
E. C. Cocke (1948 A. A. A. S. Research Grant).....	65.00
Secretarial commission.....	60.00
Academy Awards (\$20, \$15, \$5).....	40.00
Programs (N. C. State Print Shop).....	27.87
A. D. Shaftesbury (expenses High School Committee).....	24.26
Ornithological Essay Award..	20.00
Waverly Press (reprints).....	17.90
Office supplies.....	14.83
Postage.....	9.72
Expenses to meeting.....	6.84
Bank charges.....	4.08
Total.....	806.50
Bank balance.....	45.92
Cash on Hand.....	20.00
	<hr/>
	872.42

Savings Account

Balance July 1, 1948.....	66.53
---------------------------	-------

Total Assets

Checking account and cash on hand.	65.92
Savings account.....	66.53
U. S. Savings Bonds (maturity value).....	300.00
Total	432.45

On the first day of July, 1948, we, the undersigned, have examined this account and found it to be correct.

B. B. BRANDT
G. HOWARD SATTERFIELD
JOHN A. YARBROUGH, *Chairman*.

REPORT ON ACADEMY CONFERENCE AT THE CHICAGO MEETING OF THE
AMERICAN ASSOCIATION FOR THE ADVANCEMENT OF SCIENCE (1947)

Forty-three delegates from twenty-seven state academies, with the backing of half a dozen honorable visitors, wrestled helpfully with the problem of stirring the academies to aggressive activity in a science-conscious but unscientifically informed world. Skirting phases of academy-sponsored lectures or other contacts with the adult public, the burden of the discussions dwelt upon calling out latent scientific talent in high school students by the offering of significant prizes (usually scholarships to college) for high score in aptitude tests or other types of activity which would detect or forecast probable excellency in the continued study of science. The Alabama Academy surpasses all others in this phase of service according to a simple analysis of the report presented by delegate Paul Reynolds. The Alabama Academy of Science has enlisted the Alabama Chamber of Commerce to the extent of \$9,500 a year for college scholarships to five (four white and one colored) promising pupils in Alabama high schools. The annual value of these awards ranges from \$500 to \$1200. They extend for four years, provided the student continues to make good. No strings seem to be tied to the offer. The Academy is responsible for selection of the grantees and for this purpose has used the well known Westinghouse Science Talent Aptitude Test papers.

In this connection it is to be noted that of this year's 40 winners in the Westinghouse Science Talent search throughout the secondary schools of America, not one winner is from the South. Of the 260 who received honorable mention, only one out of a dozen is from our section of the country. Specifically this year's 260 pupils who received honorable mention include: one from Alabama, two from Florida, three from Georgia, one from Kentucky, five from Maryland, three each from Tennessee and Texas, and four from Virginia. The very pertinent question in this connection may be phrased, "What long-focus view and action, if any, should the North Carolina Academy of Science take concerning improvement of the quality of science teaching in the secondary schools of North Carolina?" Let this interrogatory sentence be re-read and pondered. We cannot ignore the query with a clear conscience. A windfall of dollars would

not automatically or easily convert itself into the desired result. Surely this intellectual organization has a responsibility and can work out a solution.

The conference discussions were led by Drs. A. R. Middleton of the Kentucky Academy, J. W. Cole of Virginia, H. A. Webb of Tennessee, J. W. Thomson of Wisconsin, and G. W. Prescott of Michigan. Dr. Thomson urged cooperation and friendly interchange of representatives between neighboring academies. Dr. Harlow Shapley, president of the A. A. A. S., in frank reply to some of the remarks by the gentleman from Kentucky, stated that the selection of scientific talent in the secondary schools of his state, Massachusetts, "Is in about as neglected a state as it is in Kentucky."

Prof. H. W. Michaud of Illinois presided over the meeting. Dr. A. O. Weese of Oklahoma was elected chairman and Dr. A. R. Middleton of Kentucky was elected secretary of the conference for 1948.

MILTON L. BRAUN.

The above report was accepted as information by the Academy.

REPORT OF THE LIFE MEMBERSHIP COMMITTEE

The committee, consisting of J. H. Ferguson, G. C. Mackie, G. H. Satterfield, and O. C. Bradbury, in accordance with the rules governing life membership which were established in 1943, recommended that the following persons be elected to life membership:

Archibald Henderson, Mathematics, University of North Carolina
Annie McKinnie Pegram, Mathematics, Greensboro College
E. E. Randolph, Chemical Engineering, N. C. State
Cornelia Strong, Mathematics, Woman's College, U. N. C.
B. W. Wells, Botany, N. C. State

The Academy unanimously elected these scientists to life membership.

REPORT OF THE RESEARCH GRANTS COMMITTEE

The committee has received complete reports on the work done by Dr. E. C. Cocke and Mr. T. L. Quay. These indicate that the grants for 1947 were wisely and profitably used. The committee, now consisting of B. W. Wells, G. R. MacCarthy, W. O. Puckett, and E. C. Cocke received three applications for the 1948 A. A. A. S. grant. After due consideration by correspondence it was decided to present the grant to Dr. E. C. Cocke of Wake Forest. Dr. Cocke will use the grant of \$65 for partial defrayment of expenses incurred in a study of the Myxophyceae of North Carolina.

G. R. MACCARTHY
B. W. WELLS
O. C. BRADBURY
W. E. SPEAS, *Chairman.*

REPORT OF THE POTEAT AWARD COMMITTEE

The committee met at noon on Saturday, May 8, 1948, and discussed at length the difficult problem of selecting a meritorious paper presented this year. After

due consideration it was decided to grant the award to Dr. H. Grady Britt for his excellent paper, "Gametogenesis and fertilization in the digenetic trematode, *Pneumatophilus variabilis*."

LEWIS E. ANDERSON

REINARD HARKEMA

WILLIS KING

WILLIAM MCGEHEE

H. V. PARK

G. W. WHARTON

MARY E. YARBROUGH, *Chairman*.

REPORT OF THE HIGH SCHOOL SCIENCE COMMITTEE

Dr. A. D. Shaftesbury, Chairman of the committee, gave an informal report of the past year's activities and outlined tentative plans for the coming year. Announcement was made at the evening meeting of the following winners of the awards and prizes:

THE ACADEMY AWARD

First prize: Biology group, Belmont High School, Miss M. T. Carswell, Teacher.
"Digestive Processes of Animals."

Second prize: George Hall, Belmont High School, Miss M. T. Carswell, Teacher.
"Rocks of North Carolina."

Third prize: Chemistry Class, Charlotte Central High School, Miss Mary E. Morgan, Teacher.
"Blue Printing."

BIOLOGICAL EXHIBITS AWARD

(*Carolina Biological Supply Company*)

First prize: Dave Adams, Charlotte Central High School, Miss Mary E. Morgan, Teacher.

"Preserved Mammals of Charlotte."

Second prize: Bill Cromer, Charlotte Central High School, Miss Mary E. Morgan, Teacher.

"Preserved Botanical Specimens."

Third prize: Ralph Widner, Camp Lejeune School, Camp Lejeune, N. C., H. C. Beemon, Supervising Principal.

"Microscopic Slide Collection."

ORNITHOLOGICAL ESSAY AWARD

First prize: Minerva Rae Waters, Pantego High School, N. R. Vincent, Principal.
"And They Earned Their Merit Badges, Too!"

Second prize: Charles Kiser, High School Division, Warren Wilson Junior College, Swannanoa, Henry W. Jensen, Dean.

"Birds on Our Campus."

Third prize: Frances Ann Radcliffe, Pantego High School, N. R. Vincent, Principal.

"Birds of My Home Community."

FORESTRY ESSAY AWARD

First prize: Bobby Whisnant, Spencer High School, Mrs. Elizabeth C. Kelly, Teacher.

"Forestry."

Second prize: Anne Dixon, Belmont High School, D. M. Mann, Principal.

"Growth and Care of Pines."

Third prize: Norma Lee Odom, Princeton High School, S. C. Woodard, Principal.

"Forestry on Utilization."

REPORT OF THE CONSERVATION COMMITTEE

The Conservation and Wise Use of North Carolina's Forests Must Receive Greater Emphasis

All too few people in North Carolina realize that our state ranks among the first seven leading states in lumber production and that its volume of lumber produced during several recent years has placed it fifth among all the states, being surpassed only by the Pacific Coast and one other southern state. When the great volume of timber used by other wood-using industries is added to that which goes into lumber, the forest-products industries of the state assume the proportions of "big business." Anything short of adequate protection and most efficient use of the resource upon which these industries are based would seriously impair the economic and social future of the entire state.

For over 200 years the forests of North Carolina have been "mined." Until recently forest products were abundant and cheap and vast areas of forest were destroyed by fire, insects, disease, and cutting without regard for restocking them. The extravagant use of this forest heritage contributed largely to maintaining our high standards of living. But the pioneer belief that plows would surely follow axes was not always borne out, for many areas unsuited for agriculture were denuded of their forests even though they could best serve the state and nation for timber growing.

A change in the attitude of the forest-products industries toward the source of their materials and of land owners toward their forests is under way. Within the last decade forest management has come to the fore, tending toward stable forest industries in place of migratory mills and plants, developing a permanent interest in forest lands, and the growing of new timber crops on a permanent basis instead of looking for additional virgin forests to cut. This evolution has really just gotten off to a good start and some years will be required to bring all forest lands in North Carolina under adequate protection and wise use.

Without an intensive and effective program of resource-use education, this development is likely to lag. It is too much to expect that under the old, commonly accepted American system of free enterprise, in which the economic law

of supply and demand and immediate financial returns rather than deferred income are dominant factors, reasonably good forestry will be practiced voluntarily without such education. The alternative is some reasonably effective and yet practicable form of public supervision or control over cutting on privately owned forest lands in order to keep them continuously productive of future timber crops.

If the wood-using industries are to remain permanently in business, and expansion rather than contraction of these industries is anticipated, the necessary steps must be taken to build up the annual forest growth, in the most useful sizes and species, to nearer the productive capacity of the land.

To bring about this objective, adequate fire protection must be considered first of all as a prerequisite to timber growing, and the efforts of public and private fire control agencies must be strengthened and given whole-hearted support. Forest fire control should be extended and expanded to a state-wide basis. Only about three-fourths of the counties of the state have organized fire control and most of this is inadequately financed. Education at all age levels should be intensified with greater emphasis on community responsibility and landowner participation in the fire control effort toward the primary objective of reducing vastly the number of fires occurring and the area burned.

Even though adequate control of forest fires is the first essential requirement for successful timber cropping, it alone will not necessarily keep forest lands continuously productive. Next to fire protection, the future of the forests of North Carolina depends on good cutting practices. Cutting methods that will assure perpetuation of well-stocked stands of desirable timber species should be more widely practiced. Many tree species now infrequently utilized, particularly hardwoods, should be cut and marketed. This may involve a more extensive development of wood conversion or secondary wood-using industries, possibly at the expense of lumber production. More careful attention must be given to cutting practices on privately owned forest lands to guard against stripping them without regard to their future productivity. Measures such as these will go far toward making the forests of North Carolina a growing permanent natural resource, capable of supporting more forest products industries and a larger part of the state's population. The greater natural wealth thus created will not only support enlarged permanent forest industries and supply more permanent jobs, but will also provide a wider tax base for equitable taxation.

The intensified management of forests, greatly needed in North Carolina, is highly compatible with recreational use and watershed protection. Also game production can often be realized compatibly with forest cropping.

Educational efforts in forestry began in the last decade of the 19th century and have slowly though steadily gone forward. This phase of forestry, however, is still greatly inadequate in North Carolina. A greatly enlarged and intensified campaign is urgently needed to impress the people of North Carolina with the importance of forests to their economic and social welfare. The success of forestry in North Carolina rests on a thorough understanding of the value of forests and forest products to individuals and their communities and to the state and the nation. Without such awareness by all our citizens, desirable

measures for forest management will be difficult to attain. Education to bring about this understanding should reach all segments and age groups in our population. Special effort should be made to insure fairly complete coverage in the grade and high schools through the incorporation of appropriate forest facts and materials in existing courses rather than by the introduction of new courses for this purpose. Courses giving consideration to the conservation and wise development of forest resources should be taught in all colleges in the state by teachers thoroughly trained in the subject matter.

At present the majority of our North Carolina forests are not being managed under acceptable standards of cutting, and the depletion of larger sizes and better grades and species of timber in these forests is exceeding the growth. This pronounced tendency toward forest deterioration must be fought by education of the landowners, loggers, millmen, forest industries, and the public. Education has proven its effectiveness in fire control. If the alternative of State or Federal control of cutting practice is to be escaped, education must be given sufficient men, funds, ingenuity, and effort to effect a region-wide program of sound forest management.

Many organizations, such as civic, women's, garden, and rural clubs, are taking an increasing interest in forestry. This field has been given some consideration by two special State Commissions: The North Carolina Resource-Use Education Commission and the North Carolina Education Commission. Furthermore, the North Carolina Forestry Association, recognizing the imperative need for greater educational efforts in forestry, is moving aggressively in this direction. In view of this and the fact that the General Assembly will be meeting in Raleigh during the coming winter, the Conservation Committee recommends that the Legislative Committee of the North Carolina Academy of Science be authorized and urged to include on its legislative agenda full cooperation with the North Carolina Forestry Association and other organizations and agencies in support of legislation looking toward the attainment of adequate state-wide forest fire control and wise timber cropping in North Carolina through the educational approach. The time is ripe! Let us strike while the iron is hot!

J. S. HOLMES

ARTHUR STUPKA

C. F. KORSTIAN, *Chairman*.

The above report was approved by the Academy. It was suggested that the report be published in the Journal of the North Carolina Education Association.

REPORT OF THE RESOLUTIONS COMMITTEE

The following resolutions were proposed by the committee.

Be it resolved that the North Carolina Academy of Science is deeply appreciative of the hospitality and many courtesies tendered it by the President, faculty, and student body of Davidson College; and that the Chairman of the local Committee on Arrangements is due the especial appreciation of the Academy for the part his committee is playing in the success of this, the forty-fifth annual meeting of the Academy.

Be it further resolved that a copy of this resolution become a part of the minutes of the Academy's meetings, and that copies be sent to President J. R. Cunningham of Davidson College and to Professor W. L. Porter, Chairman of the Committee on Arrangements.

Be it resolved that the North Carolina Academy of Science convey to Professor Thelma Howell, Resident Director of the Highlands Biological Laboratory, Highlands, N. C., an expression of appreciation for the work of the laboratory and for the facilities afforded by it to the biological workers interested in the flora and fauna of our mountain region.

Be it further resolved that the Academy favors expansion plans for the laboratory whereby more working space will be made available and whereby lodging and board at less than the usual resort prices may be made possible; and that it is the hope of the Academy that such plans may be carried to a successful conclusion at an early date.

Be it also resolved that the North Carolina Academy of Science strongly approve expansion plans for the U. S. Fish and Wildlife Laboratory, Beaufort, N. C., whereby a modern fireproof laboratory will be constructed for the use of scientists in many fields. Be it further resolved that a copy of this resolution be sent to the Governor of North Carolina and to each North Carolina member of Congress.

H. R. TOTTEN, *Chairman.*

The above resolutions were unanimously approved by the Academy.

The following memorial reports were presented at the meeting:

JOHN BEWLEY DERIEUX

In the death of Dr. John B. Derieux on March 17, 1948, the Academy lost one of its staunchest supporters. He joined the Academy soon after his arrival at State College in 1916 and thereafter attended practically all the meetings for thirty-two years. He served as Chairman of the Physics Section of the Academy in 1928 and in 1938 and as President of the Academy itself in 1930. In 1947 he was made a life member.

Dr. Derieux was born in 1881 in Knox County, Tennessee, where he grew up and had his early education. He entered the University of Tennessee in 1904 and received his B.S. there in 1908. While serving as an assistant principal of a rural high school in Tennessee, he also studied physics at the State University and was granted an M.S. in physics in 1914. The same year, he was appointed to a fellowship in physics at the University of Chicago and continued his studies there as fellow for two years.

After his appointment as Instructor at State College in 1916, he worked during the summers at the University of Chicago and was granted his Ph.D. in Physics in 1919. His research was directed by Dr. Millikan and his measurement of the charge on mercury droplets aided in the final measurement of the elementary

electrical charge. He published in the "Physical Review" between 1918 and 1925 three papers on this subject.

In 1920, Dr. Derieux was appointed Professor of Theoretical Physics at State College. During the following years, he identified himself with the civic and religious life of the college and community. Being musically trained and a player of wind instruments, he was leader of the choir in his Sunday School and Church for many years. His work and enthusiasm was largely responsible for the organization of a Methodist Church for the college students and the college community.

His services to State College were many. Besides teaching, he was on many important faculty committees, especially the Committee for Graduate Studies of which he was a member for twenty years. As a teacher, he was most thorough. Many are the students who failed his courses, but many there are who now are glad of the hard work they were made to do in the fundamental principles of physics and engineering. Much of his time was given to advanced courses in the teaching of which he was unexcelled. In all of his work, he showed such a personal interest in his students that he was one of the most popular of professors among the alumni as they returned for commencement exercises.

You of the Academy remember the interest with which his many papers before the Academy were received. Most were of general interest, such as "The Brown Mountain Light Investigation," 1927; "Stresses in Wire Wheels," 1931; "Highway Mirages," 1933; "Effect of Tread Upon Skidding of Tires," 1934; papers on air resistance to balls used in games, such as golf, tennis, and baseball; and many others, some seventeen in all.

We express our deep loss at the passing of this scholarly, talented, and beloved teacher and friend and regret that he was not spared to enjoy his retirement and continued friendship with those of the Academy and College who had shared with him so many pleasant experiences.

C. M. HECK.

The above memorial was approved by a rising vote.

FRANKLIN SHERMAN, JR.

Franklin Sherman, Jr., Professor of Zoology and Entomology at Clemson College, South Carolina, former President of the North Carolina Academy of Science, charter member and honorary life member, died at his home in Clemson on June 23, 1947. He is survived by his wife, the former Grace Berry; one daughter, Mrs. Grace C. Gregg; three sons, Franklin III, Dallas B., and Joseph E. Sherman.

Franklin, Jr. was born at Ash Grove, Virginia, November 2, 1877, and received his B.S.A. degree from Cornell in 1900 and his honorary M.S. degree from Maryland Agricultural College in 1912. He was an assistant in the Department of Entomology of the Maryland Agricultural Experiment Station from 1897 to 1898; Deputy Nursery Inspector of the Wisconsin Department of Agriculture, 1899; Entomologist of the North Carolina State Department of Agriculture

1900 to 1905; Professor of Entomology and Zoology, Ontario Agricultural College, 1905 to 1906; and again Entomologist of the North Carolina State Department of Agriculture 1906 to 1925; Past President of the American Association of Economic Entomologists and of the South Carolina Academy of Science.

He was a naturalist in the best sense of that word and developed the state collections in the North Carolina State Department of Agriculture and the Clemson Agricultural College. It was the writer's personal good fortune to be associated with Mr. Sherman from 1908 to 1912 when his enthusiasm for the state collection at Raleigh was at its height. He had the happy faculty of transmitting to others his boundless enthusiasm for everything in nature.

Z. P. METCALF.

The above memorial was approved by a rising vote.

CHARLES BURGESS WILLIAMS

Professor Charles Burgess Williams, agronomist for the North Carolina Agricultural Experiment Station, and one-time director, died of a heart attack at his Raleigh home, June 25, 1947. Not only was he a servant to mankind but he served God, as is evidenced by his active leadership in the Raleigh Baptist Church. His passing marked the loss of one of the nation's great leaders in agriculture. From the thousands he served have come words of commendation of a job well done. And for his Christian living and leadership, his rewards are now being realized.

Mr. Williams has left behind a name which few ever make. His strong determination to prove his convictions, even though he stood alone in many of them, proved to thousands that he was not one to be discouraged. His courage and self confidence in his own beliefs won him national recognition in his field of work.

Mr. Williams was born at Shiloh, Camden County, North Carolina, on December 23, 1871. He grew up on a general farm and attended the rural schools of that district. He later attended the Shiloh High School and Military High School at Littleton. On October 3, 1889, he entered the first class of North Carolina A. and M. College, now called N. C. State College. While a student there, he majored in agriculture and chemistry and graduated with highest honors.

In addition to being enrolled in the first class of his alma mater, Mr. Williams was captain of the first A. and M. football team. He was the first instructor to be appointed in chemistry at the college. He was the first dean of agriculture, the first head of the college agronomy department, and the first president of the Wake County State College Club.

Mr. Williams was one of the first of the trained agronomists to become interested in the growing of soybeans in this country. He became actively interested in this plant many years before it had been propagated by the U. S. Department of Agriculture. When the soybean was first planted on a very

small acreage in the eastern part of North Carolina, Mr. Williams immediately recognized its worth, and began a campaign to make it popular, a campaign that his associates say was so spectacular and interesting that it became known all over the United States.

Frank H. Jeter, Agricultural Editor at State College for the past 33 years, and one who worked closely with Mr. Williams for many years, has said this about the learned scientist, "In my opinion, Mr. Williams was the outstanding authority in the South on soils and fertilizer requirements of the various crops. He consistently worked toward the end that every foot of land in North Carolina would be surveyed before the job was completed, and it was largely due to his interest that such a large part of the survey has been completed."

In addition to his training at State College, the agronomist attended Johns Hopkins University, the National Graduate Summer School, and Ohio State University.

Mr. Williams served as assistant chemist of the North Carolina Agricultural Experiment Station, 1893-1896 and 1897-1906; director of the Experiment Station, 1907-1912; vice director of the Experiment Station, 1913; and dean of the School of Agriculture, 1917-1924.

Listed in "Who's Who in America" for many years, Mr. Williams was also a member of Phi Kappa Phi and the Pine Burr Society, both honor groups; the American Society of Agronomy; National Grange; and was a Fellow of the American Association for the Advancement of Science. He was a member of the North Carolina Academy of Science since 1902. In 1918, the "Country Gentleman," a national publication, selected him as one of seven Blue Ribbon men and women of the United States for his research work with soybeans. He was featured in 1933 in "Town and Country Review," an English magazine published in London.

The Association of Southern Agricultural Workers honored Mr. Williams at their 1941 annual meeting in Atlanta with the following citation: "Native of North Carolina, close to its soil from boyhood, graduate with highest honors in the first class of its agricultural and mechanical college, he has devoted his life to its service as chemist, agronomist, dean of agriculture, and director of its experiment station.

"He has been a leader of leaders in one of the most progressive of Southern states; has initiated and conducted experiments in a wide field of agricultural research; and is now honored for a long career of distinguished service to his state, his section, and his country."

This noted educator served State College for 53 years, the longest service of any faculty member in the history of the school. During that time, he was the author or co-author of approximately 250 articles, bulletins and books. Many of them are recognized as having lasting scientific value.

CURTIS F. TARLETON.

The above memorial was approved with a rising vote.

REPORT OF THE LEGISLATIVE COMMITTEE

This being a year that the Legislature did not meet, your committee has not been too active. We do have, however, several matters under consideration and trust that the members of the Academy will keep this in mind and will co-operate to the fullest extent when requested to help during the session of the Legislature next year.

B. C. BURGESS
H. F. PRYTHERCH
B. W. WELLS
Z. P. METCALF, *Chairman.*

REPORT OF THE NOMINATING COMMITTEE

The Committee, consisting of J. N. Couch, Chairman, M. L. Braun, and H. L. Blomquist, submitted the following nominations:

President: O. C. Bradbury, Wake Forest

Vice-President: P. J. Kramer, Duke

Member of the Executive Committee: A. F. Thiel, Woman's College U. N. C.

Member of Research Grants Committee: W. O. Puckett, Davidson; as replacement for president-elect O. C. Bradbury: E. C. Cocke, Wake Forest.

There being no nominations from the floor, the Secretary was asked to cast the ballot and declare the nominees elected. This concluded the business and the meeting was adjourned.

At 7:00 p.m. the membership enjoyed a complimentary dinner given by Davidson College. The Vice-president, Dr. A. D. Shaftesbury, presided at the evening meeting. After announcing the winners of the various high school awards, he introduced President J. R. Cunningham of Davidson College, who welcomed the Academy. This was followed by the Presidential Address by F. G. Hall, who spoke on "Physiological Adjustments to Altitude."

All of the sectional meetings were held on Saturday morning. These were well attended. The following officers were elected for the respective sections: *Biochemistry and Physiology:* Chairman, W. A. Wolff; Secretary, Mary E. Yarbrough.

Botany: Chairman, John A. Yarbrough; Secretary, Lewis E. Anderson.

Geology: Chairman, M. J. Mundorff; Secretary, E. W. Berry.

Mathematics: Chairman, W. G. McGavock; Secretary, H. V. Park.

Psychology: Chairman, William McGehee; Secretary, Marion D. Stanland.

Wildlife: Chairman, F. S. Barkalow, Jr.; Secretary, Willis King.

Zoology: Chairman, D. G. Frey; Secretary, D. S. Grosch.

The following constitute the personnel of the standing committees:

Executive: O. C. Bradbury, Paul J. Kramer, Reinard Harkema, F. G. Hall, I. E. Gray, A. F. Thiel.

Research Grants: B. W. Wells, G. R. MacCarthy, E. C. Cocke, W. O. Puckett.

Dr. O. C. Bradbury, President of the Academy for 1949, announced the appointment of the following committees:

Auditing: John A. Yarbrough, B. B. Brandt, G. H. Satterfield.

Conservation: C. F. Korstian, Arthur Stupka, R. L. Weaver.

High School: A. D. Shaftesbury, Ruby Williams, J. H. Highsmith, Mary E. Morgan, E. E. Brown.

Legislative: Z. P. Metcalf, H. F. Prytherch, B. W. Wells.

Life Membership: G. H. Satterfield, Maude Williams, John N. Couch, H. J. Oosting.

Nominating: M. L. Braun, H. L. Blomquist, R. E. Coker.

Poteat Award: L. E. Anderson, E. W. Berry, D. S. Grosch, Reinard Harkema, Willis King, H. V. Park, Marion D. Stanland, Mary E. Yarbrough.

Resolutions: O. J. Thies.

Resource Use: R. L. Weaver, H. J. Oosting.

Representative on A. A. A. S. Council: John N. Couch.

Representative to Academy Conference: Reinard Harkema.

Secretarial and Constitutional: H. R. Totten, H. L. Blomquist, Reinard Harkema.

Approximately 300 members and guests registered during the meeting.

The following papers were presented during the meeting. Those marked with an x are abstracted in these proceedings. Those marked with a † are published in full in this issue of the Journal.

GENERAL SESSIONS

Address of Welcome. DR. J. R. CUNNINGHAM, President, Davidson College.
Presidential Address: Physiological adjustments to altitude. F. G. HALL, President of the Academy.

x*The Myxophyceae of North Carolina.* E. C. COCKE, Wake Forest.

The form of the adult human stapes correlated with its functional physical characteristics. OTTO STUHLMAN, JR., U. N. C.

x*Dispersion staining.* GERMAIN CROSSMAN and W. TOM WHITE, B. & L. Optical Co.

Science needed in our education program. R. L. WEAVER, N. C. Resource-Use Commission.

Effect of wilting on permeability of roots to water. P. J. KRAMER, Duke (Read by title only).

The human side of our Museum of Natural History. F. B. MEACHAM, N. C. Museum of Natural History.

A comparative study of zygote germination in the Saprolegniaceae. A. W. ZIEGLER, U. N. C. (Published in full in this Journal 64: 13-40. 1948).

x*Heat measurements by method of cooling.* R. M. HELMS, N. C. State.

Restoration of Fish and Wildlife Laboratory at Beaufort, N. C. H. F. PRYTHERCH, U. S. Fish and Wildlife Service.

x*The inhibition of lateral buds in tobacco with growth regulators.* W. G. HARDESTY, H. T. SCOFIELD and D. B. ANDERSON, N. C. State.

BIOCHEMISTRY AND PHYSIOLOGY SECTION

x*Electrometric and colorimetric measurement of carbonic anhydrase activity.* K. M. WILBUR and N. G. ANDERSON, Duke.

Certain studies on the hydrolysis of proteins. S. L. STEELMAN and G. C. KYKER, U. N. C. School of Medicine.

x*Action of ethanolamine and ethanolamine derivatives on lipide phosphorylation.* CAMILLO ARTOM and W. E. CORNATZER, Bowman Gray School of Medicine. (This paper was presented at the 1947 meeting and is abstracted in these proceedings.)

x*The relation of chemical structure to the stimulation of lipide phosphorylation.* W. E. CORNATZER and CAMILLO ARTOM, Bowman Gray School of Medicine.

x*A possible animal analogue of familial periodic paralysis.* SUSAN GOWER SMITH, Duke School of Medicine.

x*The effect of electrolytes and sugars on the erythrocytes of the snapping turtle, Chelydra serpentina.* R. T. WINGFIELD and K. M. WILBUR, Duke.

BOTANY SECTION

The seedling of the peanut. JOHN A. YARBROUGH, Meredith.

x*An interesting epidermal cell wall.* D. E. MORELAND and D. B. ANDERSON, N. C. State.

Some observations on the structure of the egg apparatus in Torenia. LEWIS E. ANDERSON, Duke.

Comparison of algal pigments. L. A. WHITFORD, N. C. State.

x*A new genus of the Auriculariaceae.* JOHN N. COUCH, U. N. C.

An undescribed species of Papulospora parasitic on Rhizoctonia solani Kuhn. JOHN R. WARREN, Duke.

x*The marine algae of Cape Lookout, North Carolina.* L. G. WILLIAMS, U. N. C.

†*Notes on the distribution of Carya aquatica in North Carolina.* W. B. FOX, N. C. State.

The vascular flora of the olivine deposits of North Carolina and Georgia. A. E. RADFORD, U. N. C. (Published in full in this Journal 64: 45-106. 1948).

Notes on the Compositae of North Carolina. R. K. GODFREY, N. C. State.

GEOLOGY SECTION

x*Stratigraphy of the Standard Oil of New Jersey Hatteras Light Well No. 1.* LOUISE RUSSELL BERRYHILL, U. N. C.

x*Molds of slickensides in saprolite.* W. A. WHITE, U. N. C.

x*An estimate of post-triassic erosion in Piedmont, North Carolina.* J. W. HARRINGTON, U. N. C.

x*Problems of Arctic geology.* G. R. MACCARTHY, U. N. C.

†*The North Carolina Coastal Plain.* H. W. STRALEY, III, Washington, D. C.

MATHEMATICS SECTION

x*Individual instruction in College Algebra.* J. E. DANIELEY, Elon.

x*Bracket sequences.* J. M. THOMAS, Duke.

The projective-metric definition of distance. J. W. LASLEY, U. N. C.

The meaning of the interval in special relativity. ARCHIBALD HENDERSON, U. N. C. (Read by title only).

- xAutomatic electrical solution of Laplace's equation in two coordinates. P. E. GREEN, N. C. State.
- Impulse forces and the Laplace transformation. J. W. CELL, N. C. State.

PSYCHOLOGY AND CLINICAL PSYCHOLOGY SECTIONS

- xThe humane values of a science. K. L. BARKLEY, W. C. U. N. C.
- xMeasurement and prediction of student interest in natural science subjects. SAM C. WEBB, U. N. C.
- xConcept formation and emergence of contradictory relations. J. C. DIXON, U. N. C.
- The utilization of curly-cue drawings as a projective technique in the study of personality. WALLY REICHENBERG-HACKETT, Duke.
- xBasic dimensions in the description of personality. ELIZABETH DUFFY, W. C. U. N. C.
- xA picture reaction test as an exploratory device in personality study. KATHARINE M. BANHAM, Duke.

WILDLIFE SECTION

- xA report on the fish resources of lakes Jones, Salters, Singletary, White, Black, and Waccamaw. E. E. HUESKE, Wildlife Resources Commission.
- xA study of the waterfowl situation on Currituck Sound during the 1947-48 hunting season. T. S. CRITCHER, N. C. State.
- xPreliminary report on gray squirrel investigation in North Carolina. RAY ALLISON, N. C. State.
- Fur Resources in North Carolina. K. A. WILSON, Wildlife Resources Commission.
- Investigation of wildlife habitat and distribution in Wayne County. C. B. WOODHOUSE, Wildlife Resources Commission.

ZOOLOGY SECTION

- xThe cells of the fat body in haploid and diploid types of *Habrobracon*. D. S. GROSCH, N. C. State.
- xThe effects of cold on mitochondrial form in tissues of larval salamanders. H. S. ROBERTS, JR., Duke.
- xLakes of the Carolina Bays as biotic environments. D. G. FREY, U. N. C.
- xSome aspects of the development of *Axiathella mucosa* (Andrews). C. G. BOOKHOUT and E. C. HORN, Duke.
- The primary attachment of *Balanus eburneus*. G. W. WHARTON, Duke.
- xObservations on the life cycle of *Alaus oculatus* (L.) (Coleoptera: Elateridae). TOM DAGGY, Davidson.
- xGametogenesis and fertilization in the digenetic trematode, *Pneumatophilus variabilis*. H. G. BRITT, Wake Forest.
- xSimple method for preparing dry mounts of amphibian cleavage stages for class use. M. W. JOHNSON, Duke.

Abstracts:

The Myxophyceae of North Carolina. E. C. COCKE.

The study of the algae of the southeastern states has been sadly neglected. This is especially true of the Myxophyceae, on which no comprehensive work has been published for this section of the United States. Prior to 1938 only fourteen species of the blue green algae had been reported for North Carolina. Since then Drouet has published an account of ten species which were collected by Dr. Lewis D. de Schweinitz in the vicinity of Winston-Salem about 1812. In 1939 Philson reported thirty-nine new species for the state. In a master's thesis Leatherwood reported thirty-three additional species and L. A. Whitford added fifty-six more in 1942. Nine more species were added in 1946 by Blomquist and Humm, making the total number 137 species recorded for the state at the time this investigation was begun. These had been collected from only 23 counties.

During the spring and summer of 1947 the author, assisted by two students, made extensive collections throughout the state. More than 1600 samples were taken in 83 counties of North Carolina. These represented areas from the sea coast to the high mountains and thus involved a wide range of habitats. Two hundred twenty different species were collected and identified. Of this number 106 were new to the flora of the state, and several seemed new to science (*Anabaena caroliniana* sp. nov., *Anabaenopsis granulosa* sp. nov., and *Scytonema schweinitzii* sp. nov.). The number of forms new to the state summarized by families are: Chroococcaceae, 21; Oscillatoriaceae, 38; Nostocaceae, 8; Scytonemataceae, 20; Stigonemataceae, 7; Rivulariaceae, 12. This number plus 138 previously reported brings the total to 243 species of Myxophyceae now recorded for North Carolina. This investigation was made possible by the following grants and fellowships: Carnegie Grant for the Improvement of Teaching; A. A. A. S. Research Grant; Fellowship from the University of North Carolina, and the Margaret Cannon Howell Fellowship from the Highlands Biological Laboratory.

Dispersion Staining. GERMAIN CROSSMAN and W. TOM WHITE.

Microscopy in color, without the use of dyes or light filters is now a reality. Dispersion staining is the technique of introducing color to specimens under the microscope, which have not been previously stained. The color is introduced by using the ability of white light to break up into various colors of the spectrum.

Heat Measurements by Method of Cooling. R. M. HELMS.

A quantity of liquid when cooling follows Newton's Law of Cooling, even though temperatures below the freezing point are attained. When solidification starts further cooling is retarded. When solidification is completed cooling is resumed. The rate of cooling is a function of the specific heat of the substance. By comparing the rates of the substance under study with the rate of cooling of water the specific heat of the liquid substance may be found, and somewhat less certainly, the specific heat of the substance when solid.

The latent heat of fusion may be found from the rate of cooling of either the

liquid or the solid state and the time required for solidification. The time for solidification may be found analytically or graphically.

Satisfactory values were found for good heat conductors, as mercury, and for poor conductors, as sulphur. Original values for acetamide, naphthalene and urea were reported.

The method is well adapted for laboratory instruction in introductory physics courses, focusing students' attention on source of heat lost by the substance, first while cooling, then while freezing, and again in subsequent cooling.

The Inhibition of Lateral Buds in Tobacco with Growth Regulators. W. G. HARDESTY, H. T. SCOFIELD, and D. B. ANDERSON.

Attempts were made to inhibit the growth of lateral buds in field-grown plants of flue-cured tobacco by applying growth regulators to the cut-end of the stem following removal of the terminal bud. Indole acetic acid, indole butyric acid, indole propionic acid, alpha naphthalene acetic acid and the methyl ester of alpha naphthalene acetic acid were used in concentrations of 0.5%, 1.0%, 2.5%, 5.0% and 10%. Lanolin, emulsions of polyvinyl alcohol, and mineral oil were used as carriers. Six varieties of flue-cured tobacco were employed as test material. Most effective control was obtained with the methyl ester of alpha naphthalene acetic acid and with the acid itself. Indole acetic acid also gave good control in two varieties. In four of the varieties used little or no inhibition of lateral bud growth was obtained with any of the compounds. Concentrations of 5% appeared to be as effective as 10% but lower concentrations were less satisfactory. Growth of lateral buds was sometimes accelerated by the 0.5% concentrations. Lanolin and emulsions of lanolin and carnauba wax in polyvinyl alcohol were equally effective as carriers.

Electrometric and Colorimetric Measurement of Carbonic Anhydrase Activity.

K. M. WILBUR and N. G. ANDERSON.

An electrometric method for the study of the action of carbonic anhydrase as indicated by the displacement of the pH following the addition of CO₂ to a buffer solution is described. In the apparatus employed, buffer and CO₂ solutions are automatically measured and introduced with rapid mixing into a reaction vessel containing a glass electrode. Stock solutions and an arrangement for flushing the reaction vessel are incorporated with the apparatus into a single unit in an insulated chamber maintained at 0°C.

With the electrometric technique, evaluation of colorimetric methods of estimating carbonic anhydrase activity is made, and the extent of carbonic inhibition by brom thymol blue and phenol red determined. In the Roughton-Booth method, using brom thymol blue and veronal buffer, indicator inhibition is negligible and this method has been modified for research and classroom use.

The Action of Ethanolamine and Ethanolamine Derivatives on Lipide Phosphorylation. CAMILLO ARTOM and W. E. CORNATZER.

Perlman and Chaikoff have shown that a single dose of choline increases the

rate of the formation of phospholipides in the liver of rats on a high fat diet. We have recently reported that choline administration stimulates lipid phosphorylation in the intestine of rats on a low choline diet. In both liver and intestine this effect is obtained even with low fat diets, but is enhanced by the simultaneous administration of a large amount of fats.

Like choline, ethanolamine is an essential constituent of phospholipides in tissues. It may also act as a precursor of choline: the progressive methylation of ethanolamine could lead to the formation of choline through the intermediate stages of monomethyl- and dimethylethanolamine. Some evidence for the occurrence of such processes in the organism is found in several recent papers.

In the present experiments the action of ethanolamine, dimethylethanolamine, and diethanolamine on the rate of lipid phosphorylation in the liver and small intestine has been investigated with the aid of radioactive phosphorus as an indicator.

Male albino rats (100–110 gm.) were maintained for 7 days on a low choline, low fat diet. However, some of the animals received a single large dose of fat 18 hours before death. The substances tested (0.2 millimole in 1 cc.) were given by stomach tube. The rats were then injected with a solution of Na_2HPO_4 containing radioactive phosphorus, and after 6 hours they were killed by decapitation. The lipides were extracted from the liver and small intestine and their radioactivity and phosphorus content were determined as described. Animals receiving water alone were used as controls.

Results show that after administration of ethanolamine, dimethylethanolamine, and diethanolamine, the total radioactivity and the specific activity of the lipid phosphorus are higher than in the controls. This is apparent in the lipid extracted from the intestine and even more in the lipides of the liver. In both tissues the formation of phospholipides is stimulated to a degree similar or greater than that observed after the administration of an equivalent amount of choline HCl.

In a few experiments the choline-containing phospholipides of the liver have been separated with a method essentially similar to that of Taurog and associates, and the radioactivity determined in the separated fraction. From the limited number of data available, it appears that with all substances tested, the formation of both choline-containing and non-choline-containing phospholipides is accelerated. However, the increase is proportionally higher in the non-choline-containing fraction after giving diethanolamine. The same results were obtained in one experiment in which 0.2 millimoles of ethanolamine were given but not in the experiment in which this dose was doubled. When choline or dimethylethanolamine were given, the per cent increase is much greater in the choline-containing phospholipide fraction.

This investigation is being extended to other ethanolamine derivatives. At present it is apparent that ethanolamine, dimethylethanolamine, and diethanolamine all stimulate the lipid phosphorylation in the liver and intestine of rats on a choline-deficient diet. This effect is similar to that which is obtained after choline administration, and which has been correlated with the lipotropic action of choline. Dimethylethanolamine is also lipotropic. However, ethanolamine

seems to be ineffective in preventing and curing the fatty infiltration of the liver. (Aided by a grant from the John and Mary R. Markle Foundation)

The Relation of Chemical Structure to the Stimulation of Lipide Phosphorylation.

W. E. CORNATZER and CAMILLO ARTOM.

We have reported previously that the administration of a single dose of ethanolamine or of its products of partial methylation stimulated the function of total phospholipides in the liver and intestine of rats on a low protein diet. The effect of these substances is similar to that observed by others and by ourselves after feeding choline or other compounds like betaine and methionine which act as methyl donors in the body. We have now tested several other substances in order to investigate if, and to what extent, the stimulation of lipide phosphorylation could be correlated with specific chemical groups present in their molecule.

The conditions of the experiments were identical to those previously described, the rate of lipide phosphorylation being determined with the aid of radio-active phosphorus as an indicator. The substances tested (usually 0.2 millimole in one dose by stomach tube) were the following: monethyl- and diethylethanolamine, diethanolamine, triethanolamine, ethylamine, methylamine, trimethylamine, ammonium chloride, butter yellow (dimethylaminoazobenzene), glutathione, DL-serine, and inositol.

On the basis of the results of the present and previous investigations, it may be tentatively suggested that under the conditions of our experiments, a stimulating effect on lipide phosphorylation in the liver (probably also in the intestine) is observed with ethanolamine derivatives containing pentavalent as well as trivalent nitrogen in the form of primary, secondary, or tertiary amines. The presence of an alcoholic hydroxy group is probably necessary. The finding in most experiments of a stimulatory effect by ethylamine may be due to its possible conversion to ethanolamine in the body. The substitution of alkyl groups for the hydrogens linked to nitrogen seems to increase the activity, ethyl groups being as effective as methyls. The action of methyl-containing substances other than ethanolamine derivatives (such as betaine or methionine) is probably dependent on their ability to release methyl groups for the methylation of ethanolamine. However, inconsistent results were obtained with the carcinogenic dye butter yellow which is believed to contain transferable methyl groups.

Certain minor constituents of liver phospholipides (such as serine or inositol) did not show any significant effect. By and large the stimulation of lipide phosphorylation in the liver seems to have a lesser degree of chemical specificity than other known effects exerted by choline or related substances.

A Possible Animal Analogue of Familial Periodic Paralysis. SUSAN GOWER SMITH.

A paralysis has been observed in dogs on a diet low in potassium and lacking certain members of the B-complex. The paralytic process is reversed temporarily by synthetic biotin, but a more permanent cure is effected by means of potassium chloride. It is believed that a deficiency of potassium is the primary etiological cause of the syndrome.

The animal syndrome is similar to familial periodic paralysis in that preceding the final fatal progressive attack, there are varying degrees of paralysis from which the animal recovers spontaneously. The serum potassium is lowered in both instances during a severe attack, and in both, the attacks respond to potassium therapy.

Creatinuria has been observed in both the animal and the human syndromes. In both, the same muscles are affected, namely, those of the limbs, the trunk, and the neck, usually in ascending order without sensory loss or mental impairment.

Attacks may be induced artificially in the human by means of adrenalin, glucose, insulin. Attacks have been induced artificially in the dog by means of adrenalin and by a very shrill noise (in two days) produced by forcing air under pressure through a jet. Controls were given the same treatment in every case and in no instance did a control become paralyzed. The effect of the noise on a potassium-deficient dog seemed to depend on the animal's degree of depletion.

There has been no opportunity to observe the part played by heredity in our laboratory animals. In the human syndrome, Harrington (J. A. M. A. **108**: 1339, 1937) reports on incidence of 20 per cent of non-familial cases, and Gass *et al* (Medicine **27**: 105, Feb. 1948) reports in considerable detail a case which appears in all respects typical except that "No other members in his immediate or distant family suffered similarly."

The Effect of Electrolytes and Sugars on the Erythrocytes of the Snapping Turtle, Chelydra serpentina. R. T. WINGFIELD and K. M. WILBUR.

Chelydra erythrocytes exhibit a peculiar sensitivity to the absence of the calcium ion and may hemolyze in isotonic solutions lacking it. Photometric and microscopic changes in electrolyte and sugar solutions have been followed. Hemolysis occurred readily in all isotonic electrolyte solutions in the pH range 6.7-7.8, with all following relative rates at pH 7.4: $KCl > NaCl > MgCl_2$; $KCl > K_3 \text{ citrate}$; and $NaCl > Na_3 \text{ citrate}$. Below pH 6.0 hemolysis was greatly retarded. No hemolysis occurred in isosmotic $CaCl_2$ and addition of $CaCl_2$ to cells hemolyzing in $NaCl$ arrested hemolysis and caused sphering of 40-50% of ghost cells.

The hemolytic potency of isosmotic sugar solutions buffered at pH 7.4 was found to be: dextrose > sucrose > d-galactose > d-xylose, with no hemolysis in l-arabinose. Sugar hemolysis was accompanied by abnormal shape changes and disintegration of cells, whereas in Ca-free electrolyte solutions ghost cells of typical shape were found. Hemolysis in glucose was partially inhibited by $CaCl_2$, $MgCl_2$, $NaCl$, and KCl , $MgCl_2$ being least effective.

Varying degrees of agglutination occurred in unbuffered sugar solutions, the extent depending upon the particular sugar. Agglutinating action did not correlate with hemolytic potency.

An Interesting Epidermal Cell Wall. D. E. MORELAND and D. B. ANDERSON.

The outer walls of the epidermal cells of *Aucuba japonica* are made conspicuous between crossed nicols by the presence of strongly doubly refractive bands in the center of the wall. These bands or plates are usually optically negative but

may be optically positive or sometimes are both positive and negative. Staining and solubility reactions indicate that the bands are produced by the deposition of submicroscopic crystals of wax. Removal of the wax by suitable solvents eliminates the double refraction and leaves submicroscopic cavities in the cutin. Saponification of the cutin begins in these cavities and results in the disintegration of this portion of the cell wall.

A New Genus of the Auriculariaceae. J. N. COUCH.

A new genus of this family based on two species of fungi parasitic on the leaves of tropical ferns is described. The genus is named *Platycarpa* because of its flattened fruit body. One of these species collected by the late Dr. Rusby in Bolivia is described for the first time and is named *P. boliviensis*. It appears to parasitize the chlorenchyma parts of the leaf. The other species was first described as *Septobasidium polypodii* Couch. After a better understanding of this genus was obtained, however, it was recognized that this species did not belong in *Septobasidium*. It is now redescribed as *Platycarpa polypodii*. The new genus is compared with *Jola* and *Eocronartium* on mosses and *Herpobasidium* on ferns and shown to be distinct from each of these. The relationship between *Septobasidium* and *Platycarpa* is discussed.

The fungus described by Burt as *Septobasidium album* has been reexamined and scale insects have been found on the bark near the fungus with a few beneath the stroma. There is no indication, however, of any symbiosis between insects and fungus in this species. The relationship of this fungus to *Septobasidium* and to the Auriculariaceae must remain in doubt until additional material can be studied.

The Marine Algae of Cape Lookout, North Carolina. L. G. WILLIAMS.

A study extending over a period of 13 months (August, 1946, to September, 1947) was made of the marine algae growing on an artificial rock jetty at Cape Lookout, North Carolina. This locality was selected because of its remoteness from inshore water, hence having an oceanic environment, and because no previous studies have been made in this or similar habitats in localities of the Middle Atlantic Coast.

The algal flora at Cape Lookout differs markedly in species composition from that of the nearby Beaufort estuary, which has been previously studied. A comparative study of the two areas shows marked differences in (1) salinity, (2) water temperature, (3) light penetration, (4) range of tides, (5) turbidity of water, and (6) degree of contamination. It is concluded that these differences are responsible to a great extent for the differences in the floras of the two localities.

The factors of changing tides, wind, and water temperature create unusual changes in zonation throughout the year. Off-shore breezes bring lowest tides and calm seas which are responsible for the bleaching, injury and death to sublittoral species exposed during these periods. In spring and early summer typical sublittoral species occur in the intertidal zone probably because of sublittoral conditions produced by wetting from wave action.

Vertical zonation is very pronounced for many species. Generally blue-green algae and species of *Porphyra* occur as the highest growing plant life on the jetty, while species of *Dictyopteris*, *Arthrocladia*, *Peyssonnelia*, *Lithoderma* and *Grinnellia* have been found only several feet below lowest tide levels. Some species exhibit preference for sublittoral habitats but occur in the lower intertidal zones. A few species are mostly restricted to a narrow band formed between mean low tide level and low of spring tides. Many common species are found only in the lower intertidal zones. A number of species occur only as epiphytes on coarse algae, and some have been found most commonly on hydroids, sea squirts, and other sessile invertebrates.

Although the algal associations are not sharply defined, 13 communities have been recognized.

The jetty supports a permanent flora in which tropical species predominate. This is augmented by an annual winter-spring flora with northern affinities and an annual summer-fall flora with tropical affinities.

A study was also made of some of the interrelationships of sessile invertebrate animals and the algae.

One hundred thirty-two species and varieties of algae were found growing on this jetty. Fifty-four of these are new for North Carolina waters. Range limits have been extended as follows: 17 northern extensions of southern distribution, and 13 southern extensions of northern distribution. Two undescribed species were found.

An annotated systematic list of all entities identified is presented.

Stratigraphy of the Standard Oil of New Jersey Hatteras Light Well No. 1. LOUISE RUSSELL BERRYHILL.

The Hatteras Light Well No. 1 was drilled by the Standard Oil Company of New Jersey on Cape Hatteras, Dare County, North Carolina, between December 1, 1945, and July 19, 1946. Granite basement rock was encountered at 9,878 feet, and drilling was continued to a depth of 10,054 feet. A study of the well based on lithology of the samples and identification of foraminifera shows the presence of the following formations (shown with thicknesses in feet):

Pleistocene	Pamlico	142
Pliocene	Waccamaw-Croatan	130
Miocene	Upper Duplin	460
	Middle St. Marys	244
	Lower Trent	666
Eocene	Upper Castle Hayne	313
	Middle Claiborne Equivalent	477
Cretaceous	Peedec	422
	Snow Hill (?)	276
	Upper Black Creek	506
	Austin Equivalent	946
	Tuscaloosa	600
	Lower	3678

The above thicknesses are greater than those found at the outcrops of the formations. The St. Marys, Austin, and Lower Cretaceous do not appear on the surface in North Carolina.

Molds of Slickensides in Saprolite. W. A. WHITE.

Black striated surfaces found in exposures of saprolite in the piedmont have heretofore been considered to be of non-tectonic origin. Investigations by the writer show them to be molds of slickensided surfaces representing differential movement in the bedrock previous to weathering. Preservation has usually been accomplished by the emplacement of manganese oxides near the water table.

Individually these striated surfaces may represent extremely small displacement, but where large numbers of them occur with uniform orientation in the same small area, they suggest significant faulting. They may be analyzed by taking observations of the strike and dip of the surface and the orientation of the striae and plotting this information on stereographic projections.

An Estimate of Post-Triassic Erosion in Piedmont, North Carolina. J. W. HARRINGTON.

Two rotational faults were mapped along the west border of the Durham Triassic Basin. Graphic solutions of their throws indicate that Triassic sediments have been downfaulted many hundreds of feet. A restoration of the position of these sediments before faulting indicates that there has been at least 1800 feet of erosion since sedimentation time. This figure is believed to represent the correct order of magnitude for the minimum amount of erosion in North Carolina for the development of the present Piedmont surface.

Problems of Arctic Geology. G. R. MACCARTHY.

In the Arctic and Subarctic the hard rock geology is largely concealed by tundra and by glacial deposits so that, aside from limited areas that have been swept bare by the Pleistocene ice sheet, and in areas of glaciated mountains, outcrops are largely confined to stream valleys. In such regions, virtually unmapped geologically aside from narrow ribbons along certain streams, the ordinary methods of field work cannot be employed. Various geophysical methods, including the new air-borne magnetometers, may be employed to discover certain types of ore deposits and to determine geologic trend-lines by means of which scattered outcrops may be correlated. Field work is further complicated by the fact that overland transportation is all but impossible except during the winter months when not only is the terrain concealed by snow, but when temperatures as low as -70° may sometimes be encountered. The presence of many thousands of square miles of permanently frozen soil gives rise to a host of special problems in connection with railway and highway engineering, general construction work of all kinds, water supply, and the like, all of which are problems in which the arctic geologist and geophysicist are called upon for aid.

Individual Instruction in College Algebra. J. E. DANIELEY.

During the first half of the course in college algebra the class was taught by the use of a combination method which consisted of lecture and written practice or recitation. Due to the great difference in ability, age, and academic preparation as well as social and emotional maturation this method of instructing the "class as a whole" was not as effective as was desirable.

Realizing the fact that some of the students failed to comprehend and at the same time some of the students thought the material too simple and uninteresting, it was decided to put instruction on an individual basis. This was done and as a result of it there was a considerable increase in the number of students who did satisfactory work. Grades for more than sixty per cent of the class increased and more than twenty per cent of the class did work beyond that which was required of them.

Individualized instruction in algebra offers definite possibilities on the college level for the teacher who desires that all of the members of his class be challenged to do all of which they are capable.

Bracket Sequences. J. M. THOMAS.

A bracket sequence of positive integers is generated by taking the follower $x(1)$ of $x(0)$ as the greatest integer in one-half the follower of x in the sequence of natural numbers. Elementary properties of these sequences are discussed. From and after $x(k)$ every member of the sequence is unity. The positive integer $k(x) + 1$ is the number of digits when x is expressed in the binary scale. Two positive integers x, y determine a non-negative integer $q(x, y)$ characterized by the properties (i) $x(n)$ is different from $y(n)$ if n is less than q and (ii) $x(n)$ equals $y(n)$ if n is at least equal to q . Applications are found, for example, in the draws for tournaments and in the division of cells. Two players numbered x, y in the draw can meet only in round $q(x, y)$ of the tournament. Two cells of the same generation numbered x, y in a similar scheme formed part of the same cell $q(x, y)$ generations earlier.

Automatic Electrical Solution of Laplace's Equation in Two Coordinates. P. E. GREEN.

A brief resumé is given of the mathematical, graphical, and experimental methods of solving the Laplace equation in two coordinates, subject to arbitrary boundary conditions. Such solutions are useful in a number of physical applications, and the solution of one type of field problem may be applied by analogy to similar problems in other branches of physics. Attention is directed to a standard experimental attack known as the electrolytic tank method, and it is explained how such an apparatus can be modified so as to accomplish its purpose automatically and unattended. A number of plots made by such equipment are shown.

The Humane Values of a Science. K. L. BARKLEY.

In spite of its basic role in our modern culture up to date, science is not secure in the twentieth century world. The diversion of science to destructive ends

has caused some people to question whether science has humane values which outweigh its negative values and whether science can justify itself and endure. That science does have humane values no one would deny.

First, we may list the values of a science as an end in itself. These values include the pleasures and satisfactions which scientific work in itself and the making of discoveries have for the scientist. Moreover, ordinary citizens may find value in the satisfaction of their curiosities through the findings which science may offer.

The values of a science as an aid or tool in personal adjustment constitute another category. Science has aided modern man in his adjusting by freeing his mind from superstition. Through science we learn dependable methods of solving problems and we may develop an objective attitude toward the problems we face.

A third group of values is the one which has been stressed most in the past, namely, the values of a science as a source of creative invention and as a technology. These values are so many in number that we cannot give even a partial list, but they are so well known and understood they need not be emphasized here.

The fourth set of values are those which a science has as a major factor in social change. Science has changed and remolded our civilization in a thousand ways and continues to be a major factor in the creation of a new heaven and a new earth. Modern science may furnish the means by which man can continue to evolve in accord with the changing demands of a developing world.

The Measurement and Prediction of Student Interest in Natural Science Subjects.

S. C. WEBB.

This study reports the results of development of an interest inventory designed to measure and predict college students' interests in botany, chemistry, geology, physics, psychology, and zoology, and of its effectiveness in measuring and predicting interests in these subjects. Items of the test were validated separately from the standpoint of measurement and of prediction against a criterion of interest as measured by a highly reliable attitude scale. Only those items which were valid for both measurement and prediction were incorporated in the scale. Application of these tests to a different sample of 421 college students shows the median correlation between interest and test scores of students who had previously taken science courses before taking the test (index of measurement) to be 0.552, but the median correlation between interest at the end of the course with test score obtained at the beginning of the course (index of prediction) to be 0.284. Conclusions relative to the danger of using interest inventories for prediction of interests are presented.

Concept Formation and Emergence of Contradictory Relations. J. C. DIXON.

The chief problem of this investigation was to determine whether the pre-school child, upon demonstrating a functional use of the concepts *larger than* and *smaller than*, would show at the same time a behavioral recognition of the

contradiction when these two concepts are employed in a contradictory relation, i.e., whether this behavior emerged at some later time.

Behavioral recognition was defined as behavioral reaction to the contradictory relation as an unexpected event. Ability of the child to use the concepts was determined by requiring relative size judgments between pairs of geometric figures varied in such a way as to leave no consistent basis for choice except relative size. The contradictory situation was a pair of these figures, the ring-segments illusion which, upon reversal of position, results in the apparently larger segment becoming apparently smaller. Thus, a present perception is elicited which contradicts the child's immediately preceding perception and expressed judgment.

Reliability of the tests and observer agreement in judging the child's reaction to the contradictory situation were found to be satisfactory. There was sharp increase from virtually none passing either the concept or contradiction test in the age group 2-3 to approximately 50 per cent of the high and 40 per cent of the low socio-economic group passing in the age group 3-4. In the age group 5-6 all of the high and approximately 75 per cent of the low group reacted positively to the contradictory situation.

On the chief problem of investigation it was found that when the children demonstrated a functional use of the concepts employed, they also reacted to the contradictory relation posed by the ring-segments illusion in over 90 per cent of the cases. This finding is also reciprocal, i.e., when they did not show a functional use of the concepts, they did not react to the contradiction.

On the basis of this evidence it was suggested, in agreement with Spearman, that relations are essential aspects of what we mean by concepts.

Basic Dimensions in the Description of Personality. ELIZABETH DUFFY.

A major problem in any science is the selection of appropriate descriptive categories. These categories should represent single, basic aspects of phenomena which are measurable, and they should be as few as will permit the description of all functionally significant variables. The categories now employed in personality description fail to meet these requirements.

It is suggested that personality be described in terms of the *direction* and the *energy mobilization* of response. These descriptive categories, derived from the concepts of general psychology, appear to represent basic aspects of behavior. They also appear to be of universal descriptive validity, since they apply to all organisms and are valid for any culture. The particulars of description are supplied when we specify the objects, persons, or aspects of things toward or away from which the individual moves with a given intensity.

It is the contention of this paper that the proposed dimensions of behavior are functionally significant, inclusive, and measurable, and that a wide variety of trait names can advantageously be translated into these terms.

A Picture Reaction Test as an Exploratory Device in Personality Study. KATHERINE M. BANHAM.

The Picture Reaction Test is a combination of the word-association and the

thematic apperception techniques. It aims to explore personality trends and intrapsychic conflicts at a conscious and partly subconscious level, and give indications of deeper unconscious processes. Pictures have universal appeal and provide suitable test material for a wide range of subjects, including illiterates and children. The test was first devised as a means of shortening the time and reducing the labor involved in administering a full Thematic Apperception Test. The twenty Murray T.A.T. pictures are presented to the subject one at a time for five seconds each, or for ten seconds if the test is administered to a group. The subject is required to reply with the first idea which comes to his mind immediately on seeing the picture. Reaction times are recorded. This method of obtaining quick, relatively unguarded reactions serves to indicate which of the pictures are emotionally significant for the individual. These may be selected for use in a shortened form of the T.A.T.

During the first trials with this test, the quick reactions given by the subjects, and secondary associations reported after inquiry, were observed to reveal significant diagnostic information in themselves. The personality traits indicated by the picture reactions were substantiated by facts known about the actual behavior and history of the individuals concerned.

Certain types of reaction were noted, similar to the response categories isolated by Rorschach, such as whole or small detail responses, vague scenes, human movement and the like. On several of the pictures which were difficult to see, reactions to frustration were observed. These could be classified along the lines suggested by Rosenzweig, as obstacle dominant, ego-defensive, and need persistent, with a fourth type added: that of "passive affective."

Some of the "complex indicators" distinguished by Jung in the word association test, and the disturbances of the thought processes revealed in word associations and described by Rapaport, were apparent in Picture Reactions. All three stages of the perceptual process mentioned by Anna Douglas were observed in the quick reactions to pictures. The earlier "sensory" and "exploratory" stages seemed to give evidence of defense and uncertainty.

Further experimentation with the picture reaction technique is needed in order to clarify and validate the many indicators of personality traits which the test may provide.

A Report of the Fish Resources of Lakes Jones, Salters, Singletary, White, Black, and Waccamaw. E. E. HUESKE.

Eastern North Carolina contains a number of natural lakes, some of which provide good fishing while others contribute very little in this respect. In order to obtain fundamental data for fisheries management of these lakes, the North Carolina Wildlife Resources Commission conducted a survey of six of these lakes during the summer of 1947. The survey included lakes Waccamaw, White, Singletary, Jones, Salters, and Black. In this report only the fish resources are considered, the other phases of the survey being submitted in additional reports.

Lake Waccamaw was found to be the richest of lakes surveyed so far as production of fish or fishing is concerned. The species of fish that constitute most of the fisherman's creel are: white perch (*Morone americana*), black crappie

(*Pomoxis nigro-maculatus*), largemouth bass (*Micropterus salmoides*), and the bluegill (*Lepomis macrochirus*). Although there is a fairly large supply of forage fish in the lake, it should be increased in order to get better growth in the game fish. This is especially necessary in the case of the largemouth bass. In order to increase the average size of the game fish the management problem is that of increasing the forage, namely bluegills, and at the same time encourage the take of the white perch and crappie.

White Lake contains almost no forage fish and consequently, fishing is poor. The dominant species in the lake are: yellow perch (*Perca flavescens*), yellow bullheads (*Ameiurus natalis erebennus*), and largemouth bass. Growth of the largemouth bass is slightly faster than that of the yellow perch. However, in each case growth is very slow. Stomach analysis of 22 largemouth bass ranging from 2.5 inches to 13.1 inches was made and it was found that darters (*Boleosoma* sp.) formed 52% of the total number of all organisms consumed, the remainder being small sunfish, bullheads, madtoms, and crustaceans. This, of course, is not the kind of food bass prefer and satisfactory growth and reproduction cannot be expected unless the food supply is improved. The greatest attraction that White Lake offers is boating and swimming which has an undesirable effect on fishing and the spawning activities of fish. On one occasion approximately 20 new redds were made by robin (*Lepomis auritus*), all of which were destroyed during the following weekend by boating and bathing activities. Again the problem in management is to provide adequate forage, which is difficult because of the low productivity of the lake. A management plan also should include the construction of brush shelters to help fishermen catch more fish.

The remaining lakes, Singletary, Jones, Black, and Salters, are considered as a group because of similar features. These lakes provide very little sport mainly because of undesirable fish populations as well as physical and chemical conditions. Yellow bullheads and suckers (*Erimyzon* sp.) are very common and the little sunfish (*Enneacanthus gloriosus*) is the most abundant sunfish. Because of the undesirable features of these lakes it would not be practical to carry on a management program, as satisfactory results from the standpoint of fishing could not be expected.

A Study of the Waterfowl Situation on Currituck Sound during the 1947-1948 Hunting Season. T. S. CRITCHER.

The purpose of this study was to determine the species of wintering waterfowl on Currituck Sound; to estimate their total numbers and relative abundance; and to determine any factors, human, physical, or biological that affected their welfare.

The study began on December 2, 1947, and continued through the hunting season until January 12, 1948, a total of about six weeks.

Throughout this period a total of twenty-five species of waterfowl was identified. From federal census figures, it was determined that the following species showed a definite increase over the 1946-1947 season: Mallard, Black Duck, Gadwall, Canvas-back, Ring-necked Duck, American Golden-Eye, Snow Goose,

Canada Goose, Whistling Swan, and Coot. The following species, however, showed a decrease: Baldpate, Green-winged Teal, Shoveller, Pintail, Redhead, Lesser Scaup, Blue-winged Teal, Wood Duck, and Greater Scaup. The Ruddy Duck remained about the same in number for the two seasons.

Of the entire twenty-five species, Coot, Canada Goose, Canvas-back, and Ruddy Duck were by far the dominant game species, and constituted over 80% of the entire wintering population.

From hunter's kills 1,574 waterfowl were examined. Of this total, females killed exceeded males killed by over 300%. The game animals which composed the greater percentage of the hunter's bag in order listed were: Coot, Ruddy Duck, and Canvas-back.

Total population figures for the 1946-1947 season and the 1947-1948 season show an increase of approximately 87,500 waterfowl. This increase may probably be attributed to an increase in the amount of available waterfowl food in the area. Other factors such as a fly-way population shift, or an under-estimation of the numbers of young produced, could, however influence such an increase.

The numbers and types of blinds and their locations and concentrations on the sound were determined. The greater percentage of the blinds were located among the small marsh islands which occur chiefly along the eastern or outer-bank side of the sound. This concentration corresponds with the more important waterfowl feeding areas, and the effect of these blinds has a marked effect upon the waterfowl feeding activities, particularly during the times of heaviest hunting activity.

Preliminary Report on Gray Squirrel Investigation in North Carolina. RAY ALLISON.

The decline in the abundance of the gray squirrel and its importance as a game animal is offered as evidence of the need for research. Three factors which must be considered in establishing an open season are: (1) the seasonal variation in percentage of breeding animals, (2) the fluctuation in the percentage of bot-fly larvae infestation, and (3) the desire of the sportsman.

The life history, breeding habits, gestation period, number of litters per year, number of young per litter, diseases and parasites are considered. The literature relative to these subjects is reviewed.

Results of a questionnaire survey concerning the open season desired by sportsmen is reviewed: 44.4% of the individuals from mountain counties desired an open season beginning between October 15 and 31; 28.2% of those from the piedmont counties desired an open season beginning between November 15 and 31. From the coastal counties, 26% desired an open season beginning between October 15 and 31, and 21.7% between November 1 and 14.

Results of a bag check survey is presented. Of fifty-three animals checked between April 1 and October 14, 37 (69.8%) were infected. One hundred and fifteen were checked between October 15 and January 14 and 1 was infected.

Preliminary recommendations for the hunting season and recommendations for further study are made.

The Cells of the Fat Body in Haploid and Diploid Types of Habrobracon. D.S. GROSCH.

Consistently, investigation of cell size in *Habrobracon* has been based on comparisons of body surface characteristics. Because cell size differences are important in developmental metabolism and may explain relative viability of animal types, it was decided to make direct investigation of cell size in internal structure. Using an ocular micrometer to measure diameter of fat cells suspended in isotonic insect Ringer's solution and was found to be technically the most feasible method of procuring data on larger numbers of cells.

For offspring in the "two-allele" fraternities produced by the cross 25c by 25, mean fat cell diameter found for females was 0.1049 ± 0.0005 mm.; for haploid males 0.1209 ± 0.0005 mm.; and for diploid males 0.1446 ± 0.0009 mm. One hundred cells in each of ten females and of ten haploids were measured. It will be noted that haploid male size exceeds that of the females. An explanation may be based on the fact that in females relatively large abdominal volume is occupied by ovaries with contents which call for stored materials. The male gonads neither occupy as great a volume nor do the contents require storage of materials.

Diploid male fat cells are shown to be larger than those of other animal types, which is consistent with findings on cells of the body surface. Because of adjustment to fewer cells in compensation for larger cell size it was not always possible to find 100 free-floating cells in diploid male preparations. Furthermore, during the course of the experiment only six diploid males were procured which met requirements on which valid comparison of cell size could be made. These requirements were that animal size be identical and that measurements be made within an hour of the animal's emergence. Grosch (1948) demonstrated a relation between animal size and cell size. In preliminaries of the present experiments it was observed that soon after emergence fat cell size began to decrease presumably due to utilization of stored food by the animal.

The Effects of Cold on Mitochondrial Form in Tissues of Larval Salamanders. H. S. ROBERTS.

Larvae of *Ambystoma opacum* were subjected to a temperature of $1^\circ \pm 1^\circ$ C for periods ranging from 7 to 24 days. Other individuals were maintained under normal laboratory conditions except that food was withheld. Adequate control was maintained at all times. A decrease in the number of long filamentous mitochondria and an increase in the number of short forms, especially granules and spherules, was observed in liver, kidney, pancreas, intestinal mucosa, and gastric glands and mucosa. After 24 days filaments and vesicular forms were entirely absent. A study of the liver glycogen content, size changes, and mitochondrial changes, demonstrated the results to be the effect of cold and not caused by starvation resulting from failure to eat. Mitochondria changed in form by transverse fragmentation of filaments, enspherulation of long forms, and by a combination of both. A decrease in total mitochondrial material occurred in

the kidney but not in other tissues. Changes in mitochondrial distribution occurred in liver and gastric glands. The basis of these changes is not known but is suggested to involve possibly a change in chemical composition, physical organization, or both. The changes described are believed to be essentially similar to those resulting from a wide variety of experimental techniques and not a unique response to lowered temperature. Mitochondria of smooth muscle and cartilage were not observed to change in form. It is suggested that this resistance may be associated with a difference in chemical composition of the mitochondria. It is concluded that mitochondrial change in form is the expression of the alteration of normal cellular metabolism.

Lakes of the Carolina Bays as Biotic Environments. D. G. FREY.

The lakes of the Carolina Bays are similar in their morphometrical characteristics in consequence of their common mode of origin. Filling in by growth of bay vegetation has occurred most rapidly at the northwest end, so that all the lakes are displaced towards the southeast end of the bays in which they occur. The deepest portions of the lakes likewise tend to be near the southeast end, sometimes within 1000 feet of shore. All the lakes, including Waccamaw, the largest one studied, are less than 14 feet in maximum depth, and hence are too shallow for permanent thermal stratification in summer. Black Lake, the shallowest, has a maximum depth of only 7.1 feet. Only three types of bottom deposits occur—sand, fibrous peat, and pulpy peat—with sand occurring around the edge on the northeast, east, and south sides, fibrous peat along the edge at the northwest side, and pulpy peat occupying the major portion of the deep water. The deep water sand was the most productive of bottom organisms.

Drainage patterns have made all the lakes except White Lake brown in color. In this lake the swamp water can drain away from the lake, instead of having to pass through it to leave the basin. Rather highly dissociated acids have lowered the pH to less than 5 in all cases except Waccamaw, where the lime of the Waccamaw formation outcropping along the northeast end has raised the pH to neutrality.

These differences in color and pH have marked effects on the biota and productivity. Waccamaw is the only lake with an extensive mollusc population of both clams and snails; the other lakes have just one small species of ancyloid snail. White Lake is the only lake with extensive growths of vegetation in deep water, and as a result the oxygen content of the bottom water is greater than that of the surface water, just the reverse of the situation in the other lakes. Of 27 species of fish collected in the lakes, Waccamaw had 23 species, White 17 species, Jones, Salters, and Singletary each 12 species, and Black only 8. Predacious species predominated, and populations in general were small. Waccamaw with the most diverse fauna of the group likewise has the best fishing. Photosynthesis and hence productivity is greatly curtailed by the brown color of the water and by minimal quantities of plant nutrients flowing in from the small watersheds.

Some Aspects of the Development of Axiothella mucosa (Andrews). C. G. BOOKHOUT and E. C. HORN.

During the summer of 1947 collections of developmental stages of the polychaete bamboo-worm, *Axiothella mucosa*, from egg to late larval stages were made at Beaufort, North Carolina, for the purpose of studying thoroughly the external and internal changes which occur during development. Eggs are emitted in globular, jelly egg masses at dawn during the breeding season which extends from the last part of April to September.

Jelly masses containing uncleaved eggs were brought into the laboratory and followed throughout their development with the jelly mass and during tube-building stages. Cell lineage from the uncleaved egg to the 24-cell stage revealed some minor variations from the classical studies on Nereis. The larvae are modified for life within the jelly mass and differ from the typical free-swimming trochophores of many Polychaetes. While in the jelly they feed on the diatoms which flourish there from the time of egg deposition. Setigerous segments are added approximately daily from the third day onward until eleven setigerous segments are formed. At this time (10 to 12 days) the larvae emerge from the jelly masses and immediately start to build sand tubes. Coincident with this transition, growth lags and there is an interruption in the formation of new setigerous segments until approximately 25 days. Thereafter new segments are added at a more gradual rate than before until the full adult complement of eighteen setigerous segments is attained at about 50 days.

The Primary Attachment of Balanus eburneus. G. W. WHARTON.

A study of the primary attachment mechanism of the cyprid, *Balanus eburneus*, reveals that two methods are employed: one, a vacuum cup attachment, and two, an attachment by means of cement. A consideration of the forces involved in holding the cyprid to a surface suggests that there is little hope of developing antifouling surfaces that can prohibit the attachment of cyprid barnacles.

Observations on the Life Cycle of Alaus oculatus (L.) (Coleoptera: Elateridae).

TOM DAGGY.

Conclusions drawn from collection data indicate that *Alaus oculatus* (L.) females lay eggs on recently dead trees, that the young larvae excavate burrows beneath the closely fitting bark, and that as the trees decay the larvae burrow deeper into the wood. The larval period probably lasts not less than three years and possibly as much as four to six years. Pupation occurs in cells excavated in wood in the late summer or fall. The transformation usually occurs in the fall but may be delayed until spring. *A. oculatus* inhabits hardwoods only, while *A. myops* (Fab.) is said to be associated solely with conifers.

Gametogenesis and Fertilization in the Digenetic Trematode, Pneumatophilus variabilis. H. G. BRITT.

All phases of the maturation of the male sex cells in *Pneumatophilus variabilis* occur in the testes. From a single cell, the primary spermatogonium, 32 sperm

are produced. There are three spermatogonial divisions followed by two maturation divisions. The cells remain attached, following each division, resulting in a rosette-like structure consisting of 32 spermatids. The nucleus of each spermatid gives rise to a thread-like sperm. The cytoplasm does not enter into the formation of the sperm but remains in the testes as a "residual cytoplasmic mass."

Maturation of the female sex cells begins in the ovary and is completed in the upper portion of the uterus, after the entrance of the sperm and the formation of the shell. The prophase stage, which takes place in the ovary, is followed by a diffuse period which continues until the oocyte enters the ootype, where it receives the sperm. Two polar bodies are produced, neither of which was seen to divide, and a single functional egg.

The egg and sperm pronuclei do not fuse prior to the formation of the first cleavage spindle. The nuclear membranes of each pronucleus break down simultaneously and the chromosomes arrange themselves on a common spindle.

Pneumatophilus variabilis has a chromosome number of $2n=22$.

Simple Method for Preparing Dry Mounts of Amphibian Cleavage Stages for Class Use. M. W. JOHNSON.

Some of the difficulties encountered in handling formalin-preserved cleavage stages of amphibian material can be overcome by employing dry mounts.

Material is fixed in 10% formalin; jelly membranes removed; dehydrated by alcohol; and placed in xylene. Transparent plastic triangles (similar to those used for attaching small insects) are cut and placed on insect pins (no. 6). Cleavage stages are then removed from xylene to blotting paper, allowed to dry and then fastened to the tip of a triangle with some suitable adhesive. These dried mounts resemble small models and can be oriented in almost any position.

REINARD HARKEMA, *Secretary.*

PROCEEDINGS OF THE ELISHA MITCHELL SCIENTIFIC SOCIETY

1947-1948

458TH MEETING, OCTOBER 14, 1947

P. C. MAHALONOBIS: *Some Indian Problems from the Statistician's Viewpoint.*

459TH MEETING, NOVEMBER 11, 1947

E. H. HIATT: *Changes in Kidney Functions at Birth.*

O. K. RICE: *Burning of Rocket Powder.*

460TH MEETING, DECEMBER 9, 1947

H. J. MAGNUSON: *Development of Immunity in Experimental Syphilis.*

F. T. ROGERS: *Basic Theory of Carbon-Pile Type of Voltage-Regulator. Remarks on Non-Linear Steady-State Theory.*

461ST MEETING, JANUARY 13, 1948

J. C. ANDREWS: *Partial Hydrolysis of Human Hair.*

G. R. MACCARTHY: *Some Problems of the Sub-Arctic.*

462ND MEETING, FEBRUARY 17, 1948

R. H. WILEY: *Low Temperature Properties of Polymers.*

A. T. BRAUER: *Some Mathematical Games.*

463RD MEETING, MARCH 9, 1948

J. E. LARSH: *The Effect of Alcohol on Experimental Trichinosis in Mice.*

T. F. HICKERSON: *Wind Stresses in Tall Buildings Induced by Rotations and Deflections.*

464TH MEETING, APRIL 13, 1948

ARCHIBALD HENDERSON: *The Meaning of the Interval in Special Relativity.*

MAURICE WHITTINGHILL: *A New Way of Detecting Rare Crossovers Induced by Environmental Agents.*

465TH MEETING, MAY 11, 1948

D. G. FREY: *Political, Economic and Biological Factors Controlling the Occurrence and Abundance of Oysters in the Potomac River.*

A. W. ZIEGLER: *A Comparative Study of Zygote Germination in the Saprolegniaceae.*

The following officers were elected:

President (one year)—G. C. Kyker.

Vice-President (one-year)—W. L. Engels.

Corresponding Secretary (five years)—E. T. Browne

L. L. GARNER, Sec.-Treas.

OBSERVATIONS ON THE HELMINTHS AND ECTOPARASITES OF
THE COTTON RAT, *SIGMODON HISPIDUS HISPIDUS* SAY AND
ORD, IN GEORGIA AND NORTH CAROLINA

BY REINARD HARKEMA

*Department of Zoology, North Carolina State College,
Raleigh, North Carolina*

AND

LEO KARTMAN

*University of Hawaii, Agricultural Experiment Station,
Honolulu, Hawaiian Islands*

During the past few years the cotton rat has been used as an experimental animal in the study of filariasis. The presence of the filariid, *Litomosoides carinii* (Travassos), in the thoracic cavity of the cotton rat in Florida and Texas has facilitated the chemotherapeutic studies on filaria. However, no survey of the helminths parasitizing the cotton rat has been published. Cotton rats have been suggested also as potential carriers of sylvatic plague in Texas, since they harbor well known flea vectors (Craig and Faust, 1945). Because of their public health importance it is felt that similar studies are desirable in other localities where cotton rats are found. These rats, which frequent broomsedge fields (*Andropogon* spp.), are very abundant in the areas studied.

Studies on the parasites of the cotton rat on which this report is based were made independently by the authors. The Georgia rats were examined at Camp Gordon, Augusta, between December 5, 1945, and February 20, 1946, by the junior author. The North Carolina survey was made in Wake County during 1939-1942.

METHODS

All rats were trapped alive. The Georgia rats were weighed and measured before being chloroformed under a bell jar. They were thoroughly brushed with a stiff paint brush and then combed over white paper to collect the ectoparasites. All ectoparasites were killed in 70% alcohol. Nematodes were killed in hot 70% glycerin-alcohol, and cestodes fixed in alcohol-formol-acetic solution after relaxation in cool water and washing in physiological saline. The North Carolina rats were killed with illuminating gas and then weighed; no length measurements were taken. Illuminating gas proved to be a rapid lethal agent for the rats without killing the ectoparasites. Fleas were killed with a chloroformed camel's hair brush while other forms were killed in hot 70% alcohol. The rats were then skinned before dissection, and examination made of all organs under the binocular dissecting microscope. The fresh skin was spread out on a large white enameled tray and again examined for ectoparasites. Nematodes were killed in hot 70% alcohol and cestodes fixed in 5% formalin after relaxation in tepid water.

HELMINTHS FOUND

Georgia Survey

The general incidence of helminths in the 100 Georgia rats examined is given in Table 1. In 65 rats infected with *Schizotaenia sigmodontis* Chandler and Suttles a range of from 1 to 18 with an average of 4.6 worms per host was found. Twenty-eight rats were positive for the strobilocerci of *Taenia taeniaeformis* (Batsch) and showed a range of from 1 to 18 and an average of 5.4 cysts per host. Two per cent of the strobilocerci were found in the spleen. The nematode, *Mastophorus muris-ascaroides* Chitwood, ranged from 1 to 43 worms, with an average of 5.9 per rat in 76 rats. Of 40 rats examined for the filariid, *Lito-*

TABLE 1

Helminths infecting 100 cotton rats, Sigmodon h. hispidus, the number of hosts infected and the average number of parasites per infected rat at Augusta, Georgia

TAXONOMIC GROUP	SPECIES	NO. OF HOSTS INFECTED	AVERAGE NO. OF PARASITES PER INFECTED HOST
Cestoda	<i>Schizotaenia sigmodontis</i>	65	4.6
	<i>Taenia taeniaeformis</i> larvae*	28	5.4
Nematoda	<i>Mastophorus muris-ascaroides</i>	76	5.9

* The liver of one rat contained cestode larvae questionably determined as *Taenia lyncis*.

mosoides carinii, all proved to be negative. This parasite also was absent from the North Carolina rats. The intermediate host of this form, *Liponyssus bacoti* (Hirst), was not found on any of the rats examined in these surveys. This undoubtedly explains the absence of the filaria. The urinary bladder of eight rats was found to be infected with a nematode, probably *Trichosomoides crassicauda* (Bellingham). Unfortunately, these specimens were accidentally destroyed before identification could be made. It should be mentioned that the liver of one rat contained larvae questionably determined as *Taenia lyncis*.

North Carolina Survey

The incidence of the parasites in the 200 North Carolina rats examined is presented in Table 2. Sixty-two rats were infected with *Raillietina bakeri* Chandler with a range of from 1 to 61 and an average of 8.09 per host. It was noted that the size of these cestodes was often much greater than that given by Chandler (1942). These worms were larger, reaching a length of 410 mm. The larger specimens were always found when the number of cestodes per host was small. Whenever there was a very high infection the size of the worms compared favorably with Chandler's measurements. In view of the high incidence of infection with *R. bakeri* it is suggested that the cotton rat may possibly

be the normal host in North Carolina. This form was originally described from the tree squirrel, *Sciurus niger rufiventer*.

Hymenolepis diminuta (Rudolphi) was found in 43 of the examined rats with a range of from 1 to 40 and an average of 6.22 per host. *H. microstoma* (Dujardin) was found in 4 rats with a range of from 2 to 111 and an average of 32 per host. It should be mentioned, however, that the 111 specimens were scoleces

TABLE 2

Parasites infecting 200 cotton rats, S. h. hispidus, the number of hosts infected with each species, and the average number of parasites per infected host in Wake County, North Carolina

TAXONOMIC GROUP	SPECIES	NO. OF HOSTS INFECTED	AVERAGE NO. OF PARASITES PER INFECTED HOST
Cestoda	<i>Railletina bakeri</i>	62	8.09
	<i>Hymenolepis diminuta</i>	43	6.22
	<i>H. microstoma</i>	4	32
	<i>Andrya microti</i>	3	2
	<i>Taenia taeniaeformis</i> larvae	31	1.75
Nematoda	<i>Longistriata adunca</i>	149	36.3
	<i>Mastophorus muris-ascaroides</i>	99	10.92
Acarina	<i>Atricholaelaps glasgowi</i>	58	7.05
	<i>Dermacentor variabilis</i> (larvae and nymphs)	35	4.14
Anoplura	<i>Hoplopleura hirsuta</i>	8	1.75
Siphonaptera	<i>Ctenophthalmus pseudagyrtes</i>	3	1.66
	<i>Peromyscopsylla scotti</i>	2	1.5
	<i>Nosopsyllus fasciatus</i>	2	1.5
	<i>Epitedia wenmanni</i>	2	1.5
	<i>Stenoponia americana</i>	1	3

and indicated very recent infection. Possibly most of these would not have matured. The presence of this worm constitutes a new host and locality record.

Andrya microti Hansen was found in 3 rats with a maximum of 3 worms. The total number of specimens was 6. Identification of this form was made by Dr. Robert Rausch.

Larval *Taenia taeniaeformis* was found in the liver of 31 rats. Eight was the largest number found in any one host and more commonly only one or two cysts were present. The average number of larvae was 1.75 per host. Hawkins (1942) first recorded the appearance of this species in cotton rats.

Two nematodes, *Mastophorus muris-ascaroides* and *Longistriata adunca* Chandler, were found infecting the hosts and were more often encountered than any other helminth. *L. adunca* was found in 149 rats with a range of from 1 to 314 and an average of 36.3 per host. This worm demonstrated definite seasonal variation which will be discussed later. *M. muris-ascaroides* was found in 99 rats with an average of 10.92 and a range of from 1 to 102. Larvae were especially common during the summer months. Chitwood (1938) has discussed the taxonomy of this genus.

ECTOPARASITES FOUND

Georgia Survey

The general incidence of ectoparasites in 35 Georgia cotton rats examined is given in Table 3. Of 20 rats infested with *Rhopalopsyllus gwyni* C. Fox, a range of from 1 to 9 with an average of 2.7 fleas was found. Of 7 rats positive

TABLE 3

Species of external parasites infesting 35 cotton rats S. h. hispidus, the number of hosts infested with each species, and the average number of parasites per infested host at Augusta, Georgia

TAXONOMIC GROUP	SPECIES	NO. OF HOSTS INFESTED	AVERAGE NO PARASITES PER INFESTED HOST
Siphonaptera	<i>Rhopalopsyllus gwyni</i>	20	2.7
	<i>Stenoponia americana</i>	7	4.8
Anoplura	<i>Hoplopleura hirsuta</i>	32	18.5
		2 rats examined for mites	
Acarina†	<i>Trombicula whartoni</i> *	1	14.0
	<i>Atricholaelaps sigmodoni</i>	2	15.5

* New host and locality record indicated by Dr. J. M. Brennan.

† Several larval ticks, probably *Dermacentor variabilis*, were also found.

for *Stenoponia americana* (Baker), a range of from 1 to 21 with an average of 4.8 fleas was found. Of the Anoplura, *Hoplopleura hirsuta* Ferris ranged from 2 to 123 lice with an average of 18.5 per rat on 32 rats. Only two rats were examined for mites. Of these, 1 rat was infested with 14 specimens of *Trombicula whartoni* Ewing. This constitutes a new host and locality record. The 2 rats harboring *Atricholaelaps sigmodoni* Strandtmann showed a range of from 9 to 22. Several specimens of larval ticks were also found. Positive identification of these was impossible, but they were probably *Dermacentor variabilis* (Say). Dr. R. A. Cooley indicated that they might be *D. variabilis*, *Ixodes sculptus* Neumann or *Rhipicephalus sanguineus* (Latreille). Unfortunately circumstances prevented the rearing of the larvae to maturity.

North Carolina Survey

All of the North Carolina rats were examined for external parasites, listed in Table 2. Acarina were more common than any of the insect ectoparasites. *Atricholaelaps glasgowi* (Ewing) was found on 58 rats with a range of from 1 to 54 and an average of 7.05. Larvae and nymphs of *D. variabilis* were found on 35 rats with an average of 4.14 and a range of from 1 to 26. The louse, *Hoplopleura hirsuta*, was found on only 8 rats with an average of 1.75 and a range of from 1 to 4. Five species of fleas were found but none was numerous. Two hosts were found to harbor *Nosopsyllus fasciatus* (Bosc) with an average of 1.5 fleas. The same incidence was found for *Peromyscopsylla scotti* I. Fox and *Epitedia wenmanni* (Rothschild). One rat was found to harbor 3 specimens of *Stenoponia americana*. *Ctenophthalmus pseudagyrtes* Baker was found on three rats with an average of 1.66 fleas.

DISCUSSION

These two surveys are of considerable interest in view of the marked differences in the number of parasites encountered in each. Only one species of adult cestode, *Schizotaenia sigmodontis*, was found in the Georgia rats while four different species: *Raillietina bakeri*, *Hymenolepis diminuta*, *H. microstoma*, and *Andrya microti* were found in the North Carolina rats. *S. sigmodontis* was not found in the latter group although it had been found previously in other North Carolina cotton rats by the senior author.

The large protospirurid, *Mastophorus muris-ascaroides*, and the larvae of *Taenia taeniaeformis* were found in both groups. *Longistriata adunca*, the common helminth in the North Carolina group, was not encountered in the Georgia survey.

The filariid worm, *Litomosoides carinii*, was not found in any of the North Carolina rats nor in 40 Georgia rats specifically examined for it. It is of interest to note that the intermediate host of this worm, *Liponyssus bacoti*, was absent from all of the rats examined.

The Georgia rats were more heavily infested with external parasites although more species of fleas were found on the North Carolina rats. Both groups of rats were parasitized with *Hoplopleura hirsuta*, *Stenoponia americana*, and probable *Dermacentor variabilis*. *Rhopalopsyllus gwyni* found on the Georgia group was absent from the North Carolina rats, while the latter group harbored *Nosopsyllus fasciatus*, *Peromyscopsylla scotti*, *Epitedia wenmanni*, and *Ctenophthalmus pseudagyrtes*, none of which was found on the Georgia rats. The genus, *Atricholaelaps*, was represented by *A. sigmodoni* on the Georgia rats and *A. glasgowi* on the others. *Trombicula whartoni* was found on one rat in Georgia.

To determine if the time of the year affects the degree of helminthic infection, the North Carolina rats were grouped according to the four seasons. *Longistriata adunca*, *Taenia taeniaeformis* larvae, and *Hymenolepis diminuta* demonstrated definite seasonal differences. Analyses of variance were conducted on the data. Because of the nature of the data, a logarithmic transformation was used. (The actual transformation was $y = \log (x + 1)$ where x is the number

of parasites observed in a rat.) Table 4 gives the geometric means for seasons with their least significant differences in per cent. The data are in terms of numbers of parasites per rat examined. These means are adjusted for sex and weight. *Longistriata adunca* showed significant differences in degrees of infection between spring (with an average of 11.28 and a percentage of infection of 83.3%) and summer (2.37 per rat and 47.3% infected), summer and fall (22.19 per rat and 89.5% infected), and fall and winter (6.06 per rat and 73.5% infected). The highest infections were incurred during the spring and fall when environmental conditions were probably more favorable for the survival of the infective stage. There was no significant difference between the spring and fall infections nor between spring and winter infections.

TABLE 4

The geometric means for seasons with their least significant difference in per cent. The data are in terms of number of parasites per host examined. These means are adjusted for sex and weight. The number of hosts infected is also presented to indicate incidence

SPECIES	SPRING (MAR.-APR.-MAY)		SUMMER (JUN.-JUL.-AUG.)		FALL (SEP.-OCT.-NOV.)		WINTER (DEC.-JAN.-FEB.)		LEAST SIGNIFICANT DIFF.
	(48 hosts) Mean	No. hosts Infected	(36 hosts) Mean	No. hosts Infected	(48 hosts) Mean	No. hosts Infected	(68 hosts) Mean	No. hosts Infected	
									%
<i>M. muris-ascaroides</i>	1.805	29	1.885	22	1.655	22	1.049	26	102.4
<i>L. adunca</i>	11.280	40	2.2272	16	22.191	43	6.060	50	112.3
<i>R. bakeri</i>	0.495	14	0.702	11	1.267	17	0.629	20	113.9
<i>H. diminuta</i>	0.328	8	1.071	16	0.293	10	0.170	9	125.0
<i>H. microstoma</i>	0.142	2	0.014	0	0.027	1	0.016	1	262.9
<i>A. microti</i>	0.053	2	0	0	0.014	1	0.001	0	2810.5
<i>T. taeniaeformis</i> larvae..	0.443	19	0.029	1	0.037	2	0.114	9	120.2

Hymenolepis diminuta showed a significant difference in degrees of infection between the spring (1.07 worms per rat and 41.1% infected) and winter (0.17 per rat and 26.4% infected). It has been pointed out by Luttermoser (1936) that the population of the intermediate hosts of this cestode decreases during the cold season at Baltimore. A similar occurrence may explain seasonal variation of this worm in North Carolina.

Taenia taeniaeformis larvae infections were significantly different between spring (0.44 larvae per rat and 75% infected) and summer (0.03 and 0.5%) and between spring and fall (0.37 and 1.5%) infections.

There is a possibility that seasonal factors may have had some effect upon the sex ratio of ectoparasites recovered from the rats in Georgia. It has been shown by Cole (1945) that male *Xenopsylla cheopis* tend to out-number the female fleas on days with a mean high temperature and for females to predominate on cold days. In the survey, which was conducted during the cold season, of 20 rats positive for *Rhopalopsyllus gwyni*, the males constituted 33.3% and the females 66.7% of all specimens collected. On the other hand, of 7 rats positive for *Stenoponia americana* the males constituted 52.9% and the females 47.1% of all specimens taken.

A perusal of both sets of data suggested that the rats of low weight had the lowest incidence of helminths. To investigate the relationship between weight and parasite incidence a parabola was fitted to the North Carolina data on *Longistriata adunca*. It was found that this relationship was significant but accounted for only a small portion of the variance in parasite incidence. Parabolas were not fitted for the other helminths because of the very small portion of variance accounted for in *L. adunca*. Further study of the correlation between weight and incidence was made, using contingency tables. It was found that a greater proportion of the heavier rats was infected. Chi-square for *Mastophorus muris-ascaroides* was significant at the 5% level and the values for *L. adunca* and *H. diminuta* had respectively the following probability of obtaining a larger value of Chi-square: 0.08 and 0.12. Furthermore, among those rats that were infected, the heavier rats tended to be more heavily infected. This

TABLE 5

The correlation coefficients of several of the helminths found in 200 North Carolina cotton rats, *Sigmodon h. hispidus*

	<i>L. adunca</i>	<i>R. bakeri</i>	<i>H. diminuta</i>	<i>T. taeniaeformis</i>
<i>M. muris-ascaroides</i>0647	.2249*	.3353*	.0732
<i>L. adunca</i>0810	-.1327	.1234
<i>R. bakeri</i>			-.0086	-.0062
<i>H. diminuta</i>				-.0260

* Probability .01.

relation was not significant for any of the parasites but approached significance in the case of *M. muris-ascaroides* (probability 0.06). In this connection it is of interest to note that Luttermoser (1936) and Herman (1939) have found adult *Rattus norvegicus* to be uniformly more heavily infected with certain species of helminths than the juveniles. If weight is considered to be one of the indices of age in rats, these data may suggest that the incidence of helminths in adults tends to be higher than in juveniles. Obviously, older rats have been exposed to infection more often than younger ones.

Although the sex ratio of the hosts collected was approximately 1:1, there appeared to be a somewhat lower incidence of helminths in the males than in the females. The statistical study of the North Carolina data, however, demonstrated that in no case were sex differences or sex by season interactions significant.

Multiple infections by helminths in the North Carolina rats were common. One hundred twenty-four of the 200 rats harbored 2 or more species, with 5 species the greatest number found. Only 3 hosts harbored this many species. Twelve hosts had 4 species, 42 hosts 3 species, and 67 hosts 2 species. Only 10 rats failed to show any worms. It is interesting to note that more hosts (53) in the fall and winter were found to be negative or harboring only one species than in the other two seasons (23 hosts). Of the 100 Georgia rats examined, 51 har-

bored 2 or more species of helminths with 3 species the greatest number found. Seventeen hosts had 3 species, 34 hosts 2 species, and 35 hosts 1 species. A total of 14 hosts failed to show any helminths. These data indicate that 49% of the Georgia rats were found harboring only 1 species of helminth or entirely negative for worms.

Since all of the helminths encountered, with the exception of *L. adunca*, require an intermediate host, the correlations of incidence of the several parasites were computed. These are given in Table 5. *H. microstoma* and *A. microti* were omitted because of their very low infection rates. Interestingly, *M. muris-ascaroides* incidence was significantly correlated with that of both *R. bakeri* and *H. diminuta*, but there was no correlation between the incidences of the two cestodes. One possible explanation is that the intermediate stages of the two cestodes develop in different intermediate hosts, both of which may serve as the hosts for *Mastophorus*.

ACKNOWLEDGMENTS

Grateful acknowledgment is made to the following individuals for aid in the determination of the parasites reported from the Georgia rats: Dr. Benjamin Schwartz for the helminths; Dr. W. L. Jellison for the Siphonaptera; Dr. J. M. Brennan for the Acarina and Mr. G. F. Augustson for Anoplura and Acarina. Thanks are also due Mr. Chester E. Story, pest control foreman at Camp Gordon, Augusta, for the essential task of trapping the rats.

Assistance in the identification of the North Carolina parasites was graciously given by Mr. Allen McIntosh, Dr. John T. Lucker, Dr. E. W. Price, and Dr. H. E. Ewing, and is hereby acknowledged. Appreciation is expressed to Mr. C. I. Bunn for the collection of the rats. Many thanks are extended to Mr. Francis J. Verlinden and Dr. H. L. Lucas of the Institute of Statistics, University of North Carolina, Raleigh, for analyzing the North Carolina data.

SUMMARY

1. One hundred cotton rats, *Sigmodon h. hispidus*, examined at Camp Gordon, Augusta, Georgia, showed 3 species of helminths. The percentage incidence and the average number of parasites per infected host were as follows: *Schizontaenia sigmodontis*, 65%, 4.6; strobilocerci of *Taenia taeniaeformis*, 28%, 5.4; *Mastophorus muris-ascaroides*, 76%, 5.9.

2. Two hundred cotton rats from Wake County, North Carolina, harbored 6 species of helminths. The percentage incidence and the average number of parasites per infected host were as follows: *Raillietina bakeri*, 31%, 8.09; *Hymenolepis diminuta*, 21.5%, 6.22; *H. microstoma* 2%, 32; larvae of *Taenia taeniaeformis*, 15.5%, 1.75; *Longistriata adunca*, 74.5%, 36.3; *Mastophorus muris-ascaroides*, 49.5%, 10.92.

3. Thirty-five Georgia rats showed 5 species of ectoparasites. The percentage incidence and average number of parasites per infested host were as follows: *Rhopalosyllus gwyni*, 57.1%, 2.7; *Stenoponia americana*, 20.0%, 4.8; and *Hoplopleura hirsuta* 91.4%, 18.5. Two of these rats examined for mites showed an

incidence and average as follows: *Trombicula whartoni*, 50.0%, 14; and *Atricholaelaps sigmodoni*, 100.0%, 15.5. Several specimens of larval ticks, probably *D. variabilis*, were also found.

4. Two hundred North Carolina rats harbored eight species of ectoparasites. The percentage incidence and average number per infested host were as follows: *Atricholaelaps glasgowi*, 29%, 7.05; larvae and nymphs of *Dermacentor variabilis*, 17.5%, 4.14; *Hoplopleura hirsuta*, 4%, 1.75; *Nosopsyllus fasciatus*, *Peromyscopsylla scotti*, and *Epitedia wenmanni*, 1%, 1.5; *Ctenophthalmus pseudagyrtes* 1.5%, 1.66, *Stenoponia americana*, 0.5%, 3.

5. Seasonal variation in incidence and degree of parasitism were demonstrated for *L. adunca*, *H. diminuta*, and *T. taeniaeformis*.

6. Heavier rats tended to be more often and more heavily infected with helminths than lighter rats.

7. Multiple infections by helminths were common in the North Carolina rats with 124 hosts harboring two or more species. Fifty-one of the Georgia hosts harbored two or more species.

8. Infections of *M. muris-ascaroides* were correlated with infections of *R. bakeri*, and *H. diminuta*.

9. The North Carolina data failed to show any significant differences in infection of male and female cotton rats.

10. There was some evidence that season may have had an effect upon the sex ratio of *Rhopalopsyllus gwyni*. There were twice as many females as males found.

11. New host and locality records are established for *T. whartoni* and *H. microstoma*.

REFERENCES

- CHANDLER, A. C. 1942. Helminths of tree squirrels in southeast Texas. *J. Parasitol.* 28(2): 135-140.
- CHITWOOD, B. G. 1938. The status of *Protospirura* vs. *Mastophorus* with a consideration of the species of these genera. *Livro Jubilar Prof. Travassos III*: 115-118.
- COLE, L. C. 1945. The effect of temperature on the sex ratio of *Xenopsylla cheopis* recovered from live rats. *Pub. Health Rep.* 60(45): 1337-1342.
- CRAIG, C. F., AND E. C. FAUST. 1945. *Clinical Parasitology*, 4th ed. Lea and Febiger, Phila. 871 p.
- HAWKINS, P. A. 1942. *Sigmodon hispidus hispidus*, a new host for the strobilocercus of *Taenia taeniaeformis*. *J. Parasitol.* 28(1): 94.
- HERMAN, C. M. 1939. A parasitological survey of wild rats in the New York Zoological Park. *Zoologica* 24(3): 305-308.
- LUTTERMOSER, G. W. 1936. A helminthological survey of Baltimore house rats (*Rattus norvegicus*). *Am. J. Hyg.* 24(2): 350-360.
- WILLIAMS, R. W. AND H. W. BROWN. 1946. The transmission of *Litomosoides carinii*, filariid parasite of the cotton rat, by the tropical rat mite, *Liponyssus*. *Science* 103 (2669): 224.

A COMPARATIVE STUDY OF THE SUSCEPTIBILITY OF GUINEA PIGS AND HAMSTERS TO AN INFECTION WITH PIG ASCARIS

BY JOHN E. LARSH, JR., AND MARGARET S. GRAVATT

*Department of Parasitology, University of North Carolina,
Chapel Hill, North Carolina*

Man is infected with the large roundworm, *Ascaris lumbricoides*, by ingesting mature eggs. The larvae hatching from the eggs make a necessary lung migration before returning to the intestine to mature into adults. To demonstrate the lobular type of pneumonia produced by these larvae during migration, it is the common practice to use guinea pigs infected with pig ascaris (*A. lumbricoides suum*). This host-parasite relationship has been studied by many workers (Ransom and Cram, 1921; Kerr, 1938; and others). Since no reports were found showing the susceptibility of hamsters to infection with this parasite, it was the purpose of the present study to compare them with guinea pigs. The criterion used for susceptibility was the number of larvae recovered from the lungs of the two hosts at intervals after infection with standard doses of eggs.

MATERIALS AND METHODS

The guinea pigs used in these experiments were purchased from a nearby dealer. The hamsters were raised in the laboratory. Both were maintained on stock diets throughout the study and handled similarly.

The methods for obtaining the ascaris eggs for infecting the animals were essentially those of Kerr (*l.c.*). The vagina and terminal one-half inch of the two horns of the uterus were removed from living swine ascaris. This material was strained through a small screen (40 mesh) to remove the tissue. The eggs isolated in this way were incubated at room temperature in Petri dishes containing a small amount of one per cent formalin solution, added to suppress bacterial growth. At least 40 days were allowed for the maturation of the eggs, *i.e.*, for the larvae within them to become infective. Eggs obtained by this method tend to adhere to one another and form large clumps, so that to make accurate counts it was necessary to treat this material to liberate the individual eggs. About 25 cc. of the material were transferred to a 50-cc. round-bottomed centrifuge tube. A small amount of sea sand was added, the tube stoppered and shaken thoroughly. The tube was then placed in an upright position and after allowing the coarse material to settle, the top of the supernatant fluid (about one cc.) was removed to another tube. This process was repeated until sufficient numbers of freed eggs were obtained. These were then washed with saline to remove the formalin, and the number needed for infecting was determined by the dilution method. All the animals were infected by forcing the eggs into the stomach through a blunted 18-gauge needle attached to a one-cc. Tuberculin syringe.

At autopsy the lungs were removed and, after observing the degree of consolidation, were digested by artificial gastric juice (one per cent hydrochloric

acid and 0.7 per cent pepsin) to liberate the larvae. The tissue was placed in a small beaker of this solution (25 cc. per gram) and teased apart with needles. This was warmed to 37°C. and then incubated at the same temperature. Constant agitation of the material was provided by an electric stirrer and digestion, in all cases, was complete within one hour. Because of this rapid digestion, it is unlikely that any larvae were lost in the process (Ransom and Cram, 1921). This material was concentrated by centrifugation to a volume of approximately one cc. The larvae present were counted by use of the Scott slide observed under 30× magnification.

EXPERIMENTAL RESULTS AND DISCUSSION

In the first experiment, 14 guinea pigs, eight weeks old, and 13 hamsters of the same age were matched according to sex. Each animal was infected with 30 eggs per gram of body weight, which represents the maximum sublethal dose for guinea pigs (Kerr, 1938). Two animals from each group were sacrificed daily beginning 24 hours after infection and continuing for six days. On the seventh day one animal from each group was autopsied and the last guinea pig was killed on the eleventh day after infection. The number of larvae recovered from the lungs of each animal is shown in Table 1.

It will be noted that in the animals killed 24 hours after infection, no larvae were recovered. In the animals killed two days after infection, the hamsters had an average of 13 larvae (0.56 percentage development) whereas only one larva was found in the two guinea pigs. It must be noted that in comparing the two groups the percentage recovery is used, since the number of eggs given at infection was based on body weight. In the animals killed three days post-infection the hamsters showed a slight increase in numbers of larvae compared with those above, averaging 17.5 (0.90 per cent). The guinea pigs, likewise, showed a few additional larvae (average two, 0.02 per cent). The hamsters killed after four days showed an increased number of larvae (41.5, 2.03 per cent). The male guinea pig had 103 larvae (only 1.34 per cent of the infecting eggs) whereas the female had none. Averaging the two (51.5, 0.67 per cent) shows that the percentage recovery continued to be lower in the guinea pigs. The data obtained five days post-infection show that the hamsters harbored an average of 130 larvae, 5.23 per cent. The male guinea pig had 96 larvae (1.35 per cent) but the female harbored 1456 (20.65 per cent). Averaging these two shows that the guinea pigs had 776 larvae (11.00 per cent). Thus, they showed for the first time a higher percentage recovery than the hamsters. After six days the two hamsters averaged 86.5 (3.79 per cent), while the two guinea pigs again had a higher average (576, 7.54 per cent). In both the animals (one hamster and one guinea pig) killed after seven days there was a reduction in the percentage recovery (1.56 and 2.98). There was a further reduction of this percentage in the one guinea pig killed 11 days after infection (1.10).

The above results for the guinea pigs agree with the observations of earlier workers (Ransom and Cram, 1921; Kerr, 1938) that very few larvae reach the lungs within three days after infection. With the dosage used here, only five

worms were recovered from six animals. Kerr's work shows that during this time the worms are in the liver, and that in this organ many are walled off and destroyed. In the present work, the greatest number of larvae in guinea pigs was found after five days. As the above authors point out, it requires about seven or eight days for these larvae to mature before continuing their migration. It is, therefore, interesting that there was a striking reduction in the number found from the fifth through the seventh day (average percentage recovery for the fifth day 11.0, for the sixth day 7.5, and 2.9 per cent for the one pig killed the seventh day). This would seem to indicate that many of the larvae are

TABLE 1

Showing the numbers of Ascaris larvae in the lungs of hamsters and guinea pigs killed at intervals after infection with 30 eggs per gram of body weight

ANIMAL NO.	SEX	KILLED AFTER INFECTION	HAMSTERS			GUINEA PIGS		
			Infesting dose of eggs	No. larvae recovered from lungs	Percentage recovery	Infesting dose of eggs	No. larvae recovered from lungs	Percentage recovery
		<i>days</i>						
1	M	1	2,490	0	0.00	6,000	0	0.00
2	F	1	2,160	0	0.00	6,180	0	0.00
3	M	2	2,220	14	0.63	6,600	1	0.02
4	F	2	2,460	12	0.49	7,650	0	0.00
5	M	3	2,160	19	0.88	8,610	3	0.03
6	F	3	1,740	16	0.92	8,550	1	0.01
7	M	4	2,220	47	2.12	7,680	103	1.34
8	F	4	1,860	36	1.94	6,180	0	0.00
9	M	5	2,490	159	6.39	7,110	96	1.35
10	F	5	2,490	101	4.06	7,050	1456	20.65
11	M	6	2,340	123	5.26	8,070	56	0.69
12	F	6	2,160	50	2.31	7,620	1096	14.38
13	M	7	2,250	35	1.56	7,080	211	2.98
14	M	11				8,280	91	1.10

destroyed in the lung tissue, thus supporting Kerr's statement that this organ is important in the general picture of this resistance.

The above results for the hamsters show that a much greater number of larvae reach the lungs within three days after infection than in guinea pigs. As for guinea pigs, the greatest numbers of larvae were recovered five days after infection but the average percentage recovery for the hamsters was less (5.2 versus 11.0 for the guinea pigs). Reduction in the numbers of larvae in the lungs after five days was noted in both hosts.

Considering the time necessary for the larvae to reach the lungs of guinea pigs, the above results, showing greater numbers in these animals after four days, suggest that they are more susceptible to lung invasion than are hamsters. Thus, the average lethal dose for hamsters must be greater than the 35 eggs per gram of body weight reported for guinea pigs by Kerr (1938). It was decided to perform the following experiment in an attempt to determine this dose for

hamsters. Eighteen animals, five weeks old, were divided into six groups of three each. Each animal of group one received 30 eggs per gram of body weight; those of group two, 40; group three, 45; group four, 50; group five, 55; and those of group six, 60 eggs per gram of body weight. None of the hamsters died as a result of infection indicating that the lethal dose based on body weight is more than twice that for guinea pigs.

SUMMARY

A comparison was made between guinea pigs and hamsters as to the number of ascaris larvae found in the lungs following infection with 30 eggs per gram of body weight. Much larger numbers were found in hamsters within three days. The greatest number in both hosts was found five days after infection but the percentage recovery was higher in guinea pigs at that time. In both hosts there was a reduction in larvae after the fifth day but the guinea pigs continued to show a higher percentage recovery. This showed that the hamsters were more resistant to lung invasion.

The exact lethal dose based on body weight was not determined for hamsters but it was found to be at least twice that for guinea pigs.

REFERENCES

- KERR, K. B. 1938. The cellular response in acquired resistance in guinea pigs to an infection with pig ascaris. *Am. J. Hyg.* 27: 28-51.
RANSOM, B. H., AND CRAM, E. B. 1921. The course of migration of ascaris larvae. *Am. J. Trop. Med.* 1: 129-156.

FURTHER STUDIES IN MICE ON THE EFFECT OF ALCOHOL ON ACQUIRED IMMUNITY TO *TRICHINELLA SPIRALIS*

BY JOHN E. LARSH, JR. AND ORLIN K. FLETCHER, JR.

*Department of Parasitology, University of North Carolina
Chapel Hill, North Carolina*

The effect of alcohol on the bodily processes involved in resistance to bacterial infections has been studied by many workers. However, until recently there were few reports showing the drug's effect on resistance to parasitic infections. Alcohol has been demonstrated to inhibit the operation of both natural and acquired immunity to dwarf tapeworm (*Hymenolepis*) infections in mice (Larsh, 1945, 1946). More recently, the effect of the drug on immunity of mice to *Trichinella spiralis* has been studied (Larsh and Kent, in press). In this case, the drug given prior to infection showed no effect on natural resistance but interfered with the development of immunity to reinfections. The present study on *T. spiralis* was designed to test the effect on immunity of alcohol given to the mice subsequent to infection, *i.e.*, to determine whether alcohol can break down an immunity established by prior infections.

MATERIALS AND METHODS

The mice used were obtained from the *Hymenolepis*-free mouse colony which has been maintained for a number of years. This colony is housed in an air-conditioned room at a constant temperature of 75°F. A diet of Purina dog checkers and water is provided in excess. All of the animals used in the present work were removed from the parent colony at six weeks and divided into alcoholic and control groups on the basis of sex and approximate weight. Both groups were continued on the above diet and maintained under similar environmental conditions throughout the study. Using techniques described earlier (Larsh, 1947), a record was kept of food consumption and weight change per mouse.

EXPERIMENTAL RESULTS

In the first experiment, 13 of the above mice were selected, seven to receive the alcohol and six to serve as non-drugged controls. They each received 300 *Trichinella* larvae, forced into the stomach by means of a blunted, 18-gauge needle attached to a one-cc. Tuberculin syringe. The procedures for obtaining the larvae for infections have been described elsewhere (Larsh and Kent, in press). One week later the administration of alcohol, given as the larvae, was initiated in one group and continued for three weeks. After the first three days, a daily injection of not more than 0.39 cc. of 40 per cent alcohol was given to produce complete narcosis (Larsh and Kent, *l.c.*). The first three doses were small (0.15 cc.) to allow adjustment to the drug. Twenty-four hours after the last dose of alcohol, the alcoholic and control mice were given a challenging infection of 900 larvae to determine the degree of immunity present. A second

group of six controls of the same age was given this challenging infection to show the degree of immunity in mice not previously infected, *i.e.*, natural immunity. One week after the challenging infection, all of the mice in the three groups were

TABLE 1

Showing the numbers of Trichinella spiralis in alcoholic and control mice sacrificed one week after a challenging infection

MOUSE NO.	NO. OF ADULTS RECOVERED	RATIO TO NO. LARVAE IN CHALLENGING INFECTION (900)	NO. OF LARVAE IN MUSCLES	RATIO TO NO. LARVAE IN ONE STIMULATING INFECTION (300)
A. Alcoholic mice given a stimulating infection of 300 larvae and a challenging infection of 900 larvae				
1	426	0.47	19,600	65.3 X
2	486	0.54	60,000	200.0 X
3	506	0.57	37,000	123.3 X
4	477	0.53	45,000	150.0 X
5	586	0.65	85,000	283.3 X
6	527	0.59	80,000	266.7 X
7	703	0.78	59,000	196.7 X
Average....	530.1	0.59	55,086	183.6 X
B. Non-alcoholic mice given a stimulating infection of 300 larvae and a challenging infection of 900 larvae				
1	275	0.31	35,000	116.7 X
2	402	0.45	63,500	211.7 X
3	412	0.46	35,000	116.7 X
4	521	0.58	35,500	118.7 X
5	553	0.62	55,300	184.3 X
6	542	0.60	43,100	143.7 X
Average.....	450.8	0.50	44,567	148.6 X
C. Non-alcoholic mice given only challenging infection of 900 larvae				
1	387	0.43		
2	445	0.49		
3	487	0.54		
4	572	0.63		
5	539	0.60		
6	518	0.58		
Average....	491.3	0.55		

autopsied and counts made of the numbers of adult worms recovered from the small intestine and the numbers of larvae from the musculature. The parasites were recovered and counted by previously described techniques (Larsh and Kent, *l.c.*). These results are tabulated in Table 1.

It will be noted that there was a rather high recovery of adult worms from

mice of the three groups. It can be assumed that none of these worms resulted from the stimulating infection 35 days previously, since adult worms are said to be lost from mice within 16 days after infection (Gould, 1945). Of the larvae given in the challenging infection the average percentage which developed into adults was 58.9 for the alcoholic group (A), 50.1 for the controls given both infections (B), and 54.6 for the controls given only one, the challenging infection (C). The difference in the number of adult worms recovered from the alcoholic mice (average 530.1) and the controls of group B (average 450.8) is not statistically significant, since the observed difference is less than 2.00 times its standard error. These numbers of adults from the alcoholic and control mice represent ratios of 0.59 and 0.50 to the number of larvae given in the challenging infection. The results show, therefore, that the alcohol had not affected the resistance of the mice. There is also no significant difference in the numbers of adult worms recovered from these controls (average 450.8) and from those of group C given only the challenging infection (average 491.3), which means that the former group had not developed immunity as a result of the one stimulating infection.

There is no significant difference in the numbers of larvae resulting from the stimulating infection in the alcoholic mice and the group B controls (average 55,086 and 44,567, or average ratios to number of larvae in infecting dose, 183.6 and 148.6). In making counts of larvae, 30 days are allowed after an infection, since immature forms present prior to that time are lost in the digestion process (McCoy, 1934). Therefore, the challenging infection given one week prior to autopsy did not contribute to the numbers of larvae recovered from the two groups.

Three additional experiments were performed using procedures similar to those of experiment one above. The only difference was in the number of stimulating infections given prior to the alcohol treatment. In experiment two, the mice received two stimulating infections; in experiment three, four stimulating infections; and in experiment four, five stimulating infections. These infections were given at weekly intervals. In all cases, the alcohol treatment followed one week after the last stimulating infection, and the immunity developed as a result of these infections was challenged by 900 larvae given one week prior to autopsy. The second control group (C), given only the challenging infection, was included in each experiment as a check on the degree of natural immunity present. The results of these last three experiments are tabulated separately in Tables 2, 3, and 4.

Table 2 shows that the alcoholic mice given two stimulating infections prior to receiving the drug harbored an average of 612.5 adult worms and their controls (group B), 413.2. This difference is significant statistically, since it is more than 2.00 times its standard error. This means that these results might have occurred by chance in less than five out of 100 similar experiments. It is, therefore, evident that the alcohol had interfered with the immunity produced by two stimulating infections. It is interesting that these controls harbored significantly fewer worms (average 413.2) than the group C controls given only

the challenging infection (average 502.0), indicating that mice of the former group had developed immunity from the two previous infections. The difference in the numbers of larvae in the alcoholic mice and group B controls (average 80,175 and 71,083, or average ratios 133.6 and 118.5) is not significant. These ratios are obtained by dividing the average number of larvae recovered by the

TABLE 2

Showing the numbers of parasites in alcoholic and control mice sacrificed one week after a challenging infection

MOUSE NO.	NO. OF ADULTS RECOVERED	RATIO TO NO. LARVAE IN CHALLENGING INFECTION (900)	NO. OF LARVAE IN MUSCLES	RATIO TO NO. LARVAE IN TWO STIMULATING INFECTIONS (600)
A. Alcoholic mice given two stimulating infections of 300 larvae each and a challenging infection of 900 larvae				
1	627	0.75	85,700	142.8 X
2	564	0.63	50,000	83.3 X
3	590	0.66	91,000	153.3 X
4	669	0.74	94,000	156.7 X
Average.....	612.5	0.68	80,175	133.6 X
B. Non-alcoholic mice given two stimulating infections of 300 larvae each and a challenging infection of 900 larvae				
1	473	0.53	71,500	119.2 X
2	133	0.15	42,500	70.8 X
3	354	0.40	79,000	131.7 X
4	715	0.80	74,000	123.3 X
5	396	0.44	72,000	120.0 X
6	408	0.45	87,500	145.8 X
Average...	413.2	0.46	71,083	118.5 X
C. Non-alcoholic mice given only challenging infection of 900 larvae				
1	395	0.44		
2	450	0.50		
3	560	0.62		
4	490	0.54		
5	530	0.59		
6	587	0.65		
Average	502	0.56		

number given in the stimulating infections, in this case 600. It will be noted that the average ratios are less than in the previous experiment.

Table 3 shows the results for experiment three which are similar to those of the previous experiment. The alcoholic mice given four stimulating infections had an average of 544.6 adult worms whereas this average for their controls of group B was only 397.8. This difference is significant to about the same degree

as in the previous experiment, showing that the alcohol had approximately the same effect in interfering with immunity produced by four infections as that

TABLE 3

Showing the numbers of parasites in alcoholic and control mice sacrificed one week after a challenging infection

MOUSE NO.	NO. OF ADULTS RECOVERED	RATIO TO NO. LARVAE IN CHALLENG- ING INFECTION (900)	NO. OF LARVAE IN MUSCLES	RATIO TO NO. LARVAE IN FOUR STIMULAT- ING INFECTIONS (1200)
A. Alcoholic mice given four stimulating infections of 300 larvae each and a challenging infection of 900 larvae				
1	530	0.59	75,000	62.5 X
2	649	0.72	45,000	37.5 X
3	572	0.64	85,000	70.8 X
4	570	0.63	83,000	69.2 X
5	576	0.64	64,600	53.8 X
6	588	0.65	59,000	49.2 X
7	393	0.44	72,400	60.4 X
8	617	0.69	101,400	84.5 X
9	545	0.61	54,600	45.5 X
10	427	0.48	50,800	42.3 X
11	523	0.58	63,000	52.5 X
Average.	544.6	0.61	68,527	57.1 X
B. Non-alcoholic mice given four stimulating infections of 300 larvae each and a challenging infection of 900 larvae				
1	431	0.48	110,000	91.7 X
2	508	0.57	54,000	45.0 X
3	447	0.50	51,600	43.0 X
4	516	0.57	72,000	60.0 X
5	132	0.15	62,000	51.7 X
6	353	0.39	50,000	41.7 X
Average.....	397.8	0.44	66,600	55.5 X
C. Non-alcoholic mice given only challenging infection of 900 larvae				
1	456	0.51		
2	450	0.50		
3	575	0.64		
4	595	0.66		
5	487	0.54		
6	556	0.62		
7	562	0.62		
Average.....	525.8	0.58		

produced by two. These controls had developed immunity from the four previous infections as shown by the presence of significantly fewer adult worms (average 397.8) than were present in the controls of group C given only the

challenging infection (average 525.8). The difference in the numbers of larvae in mice of the alcoholic group and group B controls is not significant (average

TABLE 4

Showing the numbers of parasites in alcoholic and control mice sacrificed one week after a challenging infection

MOUSE NO.	NO. OF ADULTS RECOVERED	RATIO TO NO. LARVAE IN CHALLENG- ING INFECTION (900)	NO. OF LARVAE IN MUSCLES	RATIO TO NO. LARVAE IN FIVE STIMULAT- ING INFECTIONS (1500)
A. Alcoholic mice given five stimulating infections of 300 larvae each and a challenging infection of 900 larvae				
1	568	0.63	90,000	60.0 X
2	487	0.54	74,000	49.3 X
3	332	0.37	84,200	56.1 X
4	586	0.65	76,000	50.7 X
5	406	0.45	82,400	54.9 X
6	527	0.59	71,200	47.5 X
7	279	0.31	46,000	30.7 X
8	475	0.53	80,000	53.3 X
9	275	0.31	50,600	33.7 X
10	339	0.38	90,000	60.0 X
11	318	0.35	74,000	49.3 X
12	354	0.39	54,000	36.0 X
Average.....	412.2	0.46	72,700	48.4 X
B. Non-alcoholic mice given five stimulating infections of 300 larvae each and a challenging infection of 900 larvae				
1	473	0.53	54,200	36.1 X
2	578	0.64	74,000	49.3 X
3	362	0.40	145,000	96.7 X
4	224	0.25	30,000	20.0 X
5	431	0.48	72,000	48.0 X
6	397	0.44	56,000	37.4 X
7	201	0.22	42,000	28.0 X
Average.....	380.9	0.42	67,600	45.1 X
C. Non-alcoholic mice given only challenging infection of 900 larvae				
1	395	0.44		
2	575	0.64		
3	578	0.64		
4	542	0.60		
5	565	0.63		
Average.....	531	0.59		

68,527 and 66,600, or average ratios 57.1 and 55.5). The ratios, in this case, are obtained by dividing the number of larvae recovered by the number in four

stimulating infections (1200). It should be noted that these average ratios are considerably less than in the previous experiment.

The results shown in Table 4 differ from those of the two previous tables in that the number of adult worms in the alcoholic mice given five stimulating infections (average 412.2) is not significantly greater than that of their controls of group B (average 380.9). This indicates, therefore, that the alcohol did not interfere with the immunity produced by the five infections. These infections resulted in the production of immunity in the controls, since they harbored significantly fewer adult worms (average 380.9) than controls of group C given only the challenging infection (average 531.0). No significant difference is noted in the numbers of larvae in the alcoholic mice and their controls of group B (average 72,700 and 67,600, or average ratios 48.4 and 45.1). In this case, the average ratio is obtained by dividing the average number of larvae recovered by the number given in five stimulating infections (1500). These average ratios again are somewhat less than in the previous experiment.

DISCUSSION

The effect of alcohol in breaking the immunity produced in mice to *T. spiralis* infection was shown to vary with the number of stimulating infections given before alcoholization. Following a single stimulating infection of 300 larvae, no effect of the drug could be demonstrated, since the alcoholic animals and their non-drugged controls given similar infections harbored about the same numbers of parasites following reinfection. These results are similar to those reported earlier for *Hymenolepis* infection in mice (Larsh, 1946). Mice immunized by infection before receiving the drug were totally immune to reinfection as were the non-drugged controls. In the present case, however, there was no indication that the control mice had developed immunity in that their parasite burden at reinfection was similar to that of non-drugged mice of the same age not previously infected.

Following two and four stimulating infections, greater numbers of adult worms were found to result from reinfection in the alcoholic mice than in the controls. In both experiments the controls had developed immunity from the previous infections. It can be assumed that a similar degree of immunity was produced in mice of the alcoholic group, since they received the same infections before being treated with the drug. It is evident, therefore, that such alcohol treatment resulted in a partial breakdown in this immunity. Although the mechanism of this interference was not studied, it is worth pointing out that the effects of the drug in reducing food intake, in causing weight loss, etc., were similar to those reported earlier (Larsh, 1945, 1946, 1947).

Following five stimulating infections similar numbers of parasites were recovered from the alcoholic mice and controls after reinfection. Since the controls had developed immunity as a result of the previous infections, it is clear that the alcohol had not interfered with this immunity as it had in the previous experiment following four stimulating infections. The inability of the alcohol to produce interference may have had a quantitative relationship, *i.e.*, the

additional stimulating infection of 300 larvae may have resulted in an increased immunity beyond that which could be altered by the above alcohol treatment.

The numbers of larvae recovered from the alcoholic and control mice of the various experiments did not differ significantly. This shows that the alcohol had not interfered with the degree of muscle invasion. It is interesting to note that in both the alcoholic and control groups the ratios of the number of larvae recovered to the total number given in the stimulating doses show a steady decline correlated with the increased doses. This would seem to indicate that immunity was increasing and it exerted a progressively greater effect in reducing the number of larvae found in the musculature.

SUMMARY

The effect of alcohol in breaking down the immunity produced in mice to *T. spiralis* infection was shown to vary with the number of infections given before alcoholization. No effect of the drug was demonstrated following a single infection. After two and four infections, the alcoholic mice showed a breakdown in their immunity but no effect was evident after five infections. A discussion is given of the relationship between the number of previous infections and the degree of alcoholic interference with the resulting immunity.

REFERENCES

- GOULD, S. E. 1945. Trichinosis. Charles C. Thomas Co., Springfield, Illinois.
- LARSH, J. E., JR. 1945. Effects of alcohol on natural resistance to the dwarf tapeworm in mice. *J. Parasitol.* **31**: 291-300.
- 1946. The effect of alcohol on the development of acquired immunity to *Hymenolepis* in mice. *J. Parasitol.* **32**: 72-78.
- 1947. The role of reduced food intake in alcoholic debilitation of mice infected with *Hymenolepis*. *J. Parasitol.* **33**: 339-344.
- AND KENT, DONALD E. The effect of alcohol on natural and acquired immunity of mice to infection with *Trichinella spiralis*. *J. Parasitol.* (In Press).
- MCCOY, O. R. 1934. The effect of vitamin A deficiency on the resistance of rats to infection with *Trichinella spiralis*. *Am. J. Hyg.* **20**: 169-180.

A SURVEY FOR INTESTINAL PROTOZOA AMONG FOOD HANDLERS OF A UNIVERSITY DINING HALL

By JOHN E. LARSH, JR., MARGARET S. GRAYATT, AND
HAZEL B. GILCHRIST

*Department of Parasitology, University of North Carolina
Chapel Hill, North Carolina*

During the month of January, 1947, of the stools examined at the University Infirmary, a greatly increased percentage was found to be positive for various species of intestinal protozoa, including the pathogenic amoeba, *Endamoeba histolytica*. As most of these students had been eating regularly at Lenoir dining hall, the University Physician, Dr. E. McG. Hedgpeth, and the District Health Officer, Dr. O. D. Garvin, thought it advisable to make the following survey. Only five of the 58 food handlers examined were not employed at the Lenoir dining hall.

In the many surveys that have been made, there has been general agreement that the intestinal protozoa, including *E. histolytica*, are rather prevalent in the population. However, there is controversy concerning the importance of food handlers in the epidemiology of these infections. A few selected references will be considered below in the discussion of the results of the present survey.

METHODS

The protozoa living in the human intestine can be demonstrated in stool specimens if care is taken in the collection and examination of the material. For the most part, Craig's (1948) recommendations were followed here. The reader is referred to this reference for details of the various methods. As this author points out, a much larger percentage of positive infections can be demonstrated by giving a saline cathartic and examining material from a second freshly voided stool. This procedure was not adaptable to the present study, however, as the employees had to be examined during work hours. Thus an arrangement was made whereby each of them was transported to the Infirmary where a sample was obtained by saline enema and brought to the laboratory for immediate examination.

Semi-fluid and fluid stools, likely to contain only motile forms (trophozoites), were each examined by two simple saline smears and two smears prepared by Quensel's method. The latter preparations were set aside for 10-20 minutes to allow penetration of the stain; during this time the saline preparations were studied to observe the unstained material. The Quensel's method is of great value in differentiating the trophozoites of the various amoebae, since it stains the nuclear parts. It does not stain the amoebae cysts or intestinal flagellates, but these are easily seen against the blue background provided by the stain.

In the case of semi-formed and formed stools, likely to have only cysts, examinations were made by two simple saline smears and two smears stained with

D'Antoni's iodine. In addition, an iodine smear was examined of material obtained by using the simplified zinc sulphate flotation method of Otto *et al* (1941). This method, useful for concentrating cysts, was employed in an attempt to detect light infections which might be missed by the direct fecal examinations. In cases where iodine was used, the preparations were set aside for at least five minutes to allow for penetration.

The various fecal smears were examined under low power of the microscope and the entire area covered. The high dry, and rarely oil immersion, objective was used for identifying the organisms.

Culturing methods, used routinely in this laboratory as an aid in diagnosis of *E. histolytica*, were not employed as they are too time consuming for survey work.

RESULTS AND DISCUSSION

The results of this survey, involving the examination of only one stool from each of 58 food handlers (53 from Lenoir hall), are shown in Table 1. It will be noted that 39 (67.2 per cent) of the 58 individuals examined showed no intestinal protozoa by the methods employed, whereas 19 (32.8 per cent) were found to harbor one or more of these parasites. Of the common intestinal protozoa, only *Dientamoeba fragilis* among the amoebae and *Trichomonas hominis* among the flagellates were not detected. Of the 19 persons shown to harbor protozoa, 15 were positive for one species. It is important to note that 4 (6.9 per cent) of the total examined were carriers of *E. histolytica*. These were treated at the Infirmary. The table shows that three of the individuals had double infections with *Endamoeba coli* and *Endolimax nana* while one person harbored three different parasites, *E. histolytica*, *E. nana*, and *Chilomastix mesnili*. It is of interest that no stools were found positive by the zinc sulphate concentration method which had not been found positive by the direct smears. This probably can be explained by the presence of large numbers of cysts in the positive samples. As stated above, this method is of value in diagnosis when the number of cysts is too small to be detected by direct smears.

The infection rates of the present survey are low as compared with those found by other workers following three or more examinations of each person. Since it is known that repeated examinations are often necessary to detect these infections, the lower rates reported here probably were due to the necessity of limiting the examinations to one per individual. This limitation would not have been as much of a handicap in this respect had it been possible, as mentioned above, to obtain the specimens following purgation rather than after an enema.

There have been many surveys on the prevalence of intestinal protozoa in the population (McMullen and Gray, 1941; Tsuchiya and Jean, 1940; Rothman and Laskey, 1943; Anderson *et al*, 1948). The results vary considerably, due undoubtedly to a variety of factors, including conditions of the area, socio-economic status of the population, sampling techniques, etc. However, the results, all showing a high percentage infection serve to emphasize the widespread distribution of these parasites in the United States. There also have been many such

surveys limited to food handlers, but few references were found in which conditions were similar enough for comparison with the present study. The most comparable is that of Wenrich and Arnett (1942). In the first examination of their employee group of 50 persons, they reported the following. Thirty-one (62 per cent) were infected with intestinal protozoa. Of the 50 examined, 20 (40 per cent) harbored *E. nana*; 14 (28 per cent), *E. coli*; 4 (8 per cent), *E. histolytica*; 4 (8 per cent), *G. lamblia*; 2 (4 per cent), *C. mesnili*, but none harbored *Iodamoeba williamsi*. Comparing these results with Table 1, one notes a consistently lower infection rate in the present study except for *I. williamsi*. A partial explanation for this may be in the omission from the present study of permanent stained preparations, which the above authors point out added greatly to the number found positive for *E. histolytica* and *E. nana*.

TABLE 1

The prevalence of intestinal protozoa as revealed by one stool examination of 58 food handlers

	NUMBER	PERCENTAGE
Individuals examined	58	
Positive cases*	19	32.8
<i>E. histolytica</i>	4	6.9
<i>E. coli</i>	8	13.8
<i>E. nana</i>	9	15.5
<i>I. williamsi</i>	1	1.7
<i>G. lamblia</i>	1	1.7
<i>C. mesnili</i>	1	1.7

* This number includes 15 single infections, three double infections (*E. coli* and *E. nana*), and one triple infection (*E. histolytica*, *E. nana*, and *C. mesnili*).

The role of the food handler in the transmission of intestinal protozoa is not yet clear despite the many studies, especially concerning *E. histolytica*, that have been made. Some workers (Craig, 1934; Schoenleber, 1940) emphasize the danger, while others (Sapero and Johnson, 1939) minimize the danger involved. Despite the opportune position of the food handler, especially those handling uncooked foods, it is likely that the danger in transmitting these infections and others is related to the peculiar conditions of a given situation. It, therefore, would be necessary to make thorough epidemiological studies in each situation to determine the danger. Such an extensive study was not possible in the present case. The results, however, did not reveal an infection rate greater than would be expected in the general population. This, and especially in view of the local conditions, would make it seem unlikely that the food handlers had an important role in the increased number of infections observed in students just prior to the survey.

SUMMARY

Results of a survey among 58 food handlers are presented, which revealed the presence of four species of amoebae (*E. histolytica*, *E. coli*, *E. nana*, and *I.*

williamsi) and two species of flagellates, (*G. lamblia* and *C. mesnili*). These results are compared with those of certain other surveys and the suggestion offered that food handlers in the present case probably were not responsible for an increased number of infections among students.

The writers acknowledge with appreciation the cooperation of Mr. J. A. Westbrook, Sanitary Engineer, District Health Department, and certain members of the Infirmary staff in making this survey possible.

REFERENCES

- ANDERSON, H. H., JOHNSTONE, H. G., PACKER, H., YOUNG, J. D., CALLISON, M. K., HAMBL, A. S., JR., ANDERSON, J. VAN D., AND REED, R. K. 1948. Incidence and control of amebiasis and other intestinal protozoan infections in a Memphis hospital. *Amer. J. Trop. Med.* **28**: 373-382.
- CRAIG, C. F. 1934. Amebiasis and Amebic Dysentery. Charles C. Thomas. Springfield, Illinois.
- . 1948. Laboratory Diagnosis of Protozoan Diseases. Lea and Febiger. Philadelphia, Pa.
- McMULLEN, D. B., AND GRAY, J. K. 1941. The incidence of intestinal parasites in fecal samples collected in eastern Oklahoma. *Sou. Med. Jour.* **34**: 177-181.
- OTTO, G. F., HEWITT, R., AND STRAHAN, D. E. 1941. A simplified zinc sulfate levitation method of fecal examination for protozoan cysts and hookworm eggs. *Amer. Jour. Hyg.* **33**: 32-37.
- ROTHMAN, M. M., AND LASKEY, M. 1943. Survey of protozoan infection of the staff of a large general hospital. *Amer. Jour. Med. Sci.* **206**: 369-371.
- SAPERO, J. J., AND JOHNSON, C. M. 1939. An evaluation of the role of the food handler in the transmission of amebiasis. *Amer. J. Trop. Med.* **19**: 255-264.
- SCHOENLEBER, A. W. 1940. The food handler as a transmitter of amebiasis. *Amer. J. Trop. Med.* **20**: 99-106.
- TSUCHIYA, H., AND JEAN, J. T. 1940. The incidence of intestinal protozoa among freshman medical and dental students with especial reference to amebiasis. *Amer. J. Trop. Med.* **20**: 803-808.
- WENRICH, D. H., AND ARNETT, J. H. 1942. Results of a protozoological survey of food handlers at a professional school in Philadelphia, Pa. *Amer. J. Trop. Med.* **22**: 107-115.

COMMON ECTOPARASITES OF THE NORWAY RAT, *RATTUS NORVEGICUS* (ERXL.), IN NORTH CAROLINA AND THE EFFECTS OF 10 PER CENT DDT DUST TREATMENTS IN THEIR CONTROL AND UPON THE SEROLOGY OF THE RAT

BY H. F. SCHOOF¹

*North Carolina State Board of Health
Raleigh, North Carolina*

In September, 1945, the North Carolina State Board of Health in cooperation with the U. S. Public Health Service, Malaria Control in War Areas,² inaugurated a DDT-dusting program for the control of endemic typhus in the state. Initially, the program was limited to the four communities, Wilson, Wilmington, New Bern, and Clinton, where past records indicated the disease to be most prevalent. From this limited coverage, the program expanded into ten towns, many of which now participate in the expense of control.

The control measure consisted of the application of a 10% DDT dust to the rat runs and harborages of all premises, including residences, food-handling establishments, feed stores, and other businesses.

In each town, entomological procedures were carried on to determine the effectiveness of the control measures. Prior to dusting operations, a sample of the rat population was trapped and examined for ectoparasites. The pre-dust ectoparasitic index was thus obtained, and could then be compared with indices secured from post-dust samples. The initial post-dust samples were usually taken within 5-15 days after dusting occurred, with subsequent examinations being made at four to eight-week intervals. For each inspection, the same establishment and its environs (400'-500') served as trapping stations. Occasional random samples were also taken within the area as a check on whether or not the dusting crews were concentrating their efforts on the sampling premises.

To secure the ectoparasitic sample, the rat was killed by chloroform and placed on a white-enamelled pan. A fine-toothed comb was then stroked briskly through the hair, from the tail toward the head, to loosen the parasites. After combing, the rat was held by the tail and struck sharply several times with a stick to dislodge the parasites. Specimens were removed from the pan by a small brush and stored in 70% alcohol for identification.

Blood specimens were also obtained from all rats to determine, by a complement fixation test,³ (Bengtson, 1941), the percentage which had contracted a typhus infection.

¹ Entomologist, P. A. Sanitarian (R), U. S. Public Health Service.

² These activities were taken over by the Communicable Disease Center, Public Health Service, on July 1, 1946.

³ Performed under the supervision of the North Carolina State Laboratory of Hygiene by the author and biological aides.

COMMON ECTOPARASITES OF *RATTUS NORVEGICUS*

The examination of 854 specimens of *R. norvegicus* from 17 undusted communities in the state revealed that the rat louse, *Polyplax spinulosa*, was far more abundant than any other ectoparasite, comprising 74.9 per cent of the total population (Table I). Approximately 65 per cent of the rats⁴ were infested by this louse with an average of 32.2 lice being found per rat. *P. spinulosa* was most prevalent during the winter and spring seasons, the fall densities being the

TABLE I

The percentage of rats infested and the relative percentages of the principal ectoparasites recovered from combined samples of rats taken in 17 undusted towns in North Carolina. Relative percentages shown in parentheses

	DEC.-JAN.- FEB.	MAR.-APR.- MAY	JUNE-JULY- AUG.	SEPT.-OCT.- NOV.	JAN.-DEC.
<i>Xenopsylla cheopis</i> (Roths.)*.	29.7 (2.3)	28.3 (3.6)	43.0 (7.2)	47.7 (22.7)	34.5 (5.2)
<i>Nosopsyllus fasciatus</i> (Bosc.)..	22.7 (1.1)	48.4 (7.5)	46.9 (4.6)	20.1 (2.3)	33.8 (4.2)
<i>Leptopsylla segnis</i> (Schön.) .	9.2 (.7)	8.3 (1.7)	5.5 (.3)	6.5 (2.0)	7.9 (1.2)
<i>Echidnophaga gallinacea</i> (Westw.)	2.7 (.2)	4.0 (4.6)	0 (0)	13.0 (16.6)	5.3 (3.7)
<i>Liponyssus bacoti</i> (Hirst) ..	5.0 (.8)	20.6 (3.0)	43.0 (27.1)	21.1 (3.7)	17.9 (3.3)
<i>Atricholaelaps glasgowi</i> (Ewing).	3.0 (.1)	10.1 (1.0)	5.5 (.2)	4.5 (8.1)	6.3 (1.3)
<i>Echinolaelaps echidninus</i> (Berlese).....	33.2 (6.2)	17.9 (1.6)	18.0 (6.7)	36.6 (22.0)	26.9 (5.9)
<i>Polyplax spinulosa</i> (Bur- meister)	72.2 (88.5)	72.8 (76.8)	61.1 (53.5)	42.2 (22.0)	64.5 (74.9)
Number of <i>R. norvegicus</i> . . .	259	324	72	199	854
Total number of ectopara- sites	15,328	15,824	1821	3788	36,771

* *Ctenocephalides canis* (Curtis), *Ctenocephalides felis* (Bouche), *Ctenophthalmus pseudagyrtes* (Baker), *Laelaps nuttalli* (Hirst), *Eulaelaps* sp?, *Uropoda* sp?, *Chelytes* sp?, *Derma-centor variabilis* (Say) were collected in densities less than .1 per cent.

lowest. During the latter season, this species comprised 22 per cent of the total ectoparasitic population (as compared to 88.5 per cent in the winter) and was found on only 42.2 per cent of the rats.

When the flea densities are considered separately (Table II), three species, *Xenopsylla cheopis*, *Nosopsyllus fasciatus*, and *Echidnophaga gallinacea*, constitute approximately 91 per cent of the total number taken. The oriental rat flea, *X. cheopis*, showed the greatest density, being found on 36.1 per cent of the rats at an average of 2.3 fleas per rat. Seasonally, this species was most prevalent in the fall when it infested 47.7 per cent of the rat population at an average of 4.3 fleas per rat. In contrast, *N. fasciatus* was found most frequently in the spring and summer seasons with the fall densities being the lowest.

⁴ Percentage of infested rats is based upon the number of rats which yield one or more ectoparasites of the species concerned.

The stick-tight hen flea, *E. gallinacea*, during the season of its greatest density infested only 13 per cent of the rats at an average of 3.1 fleas per rat. It is of interest to note that while this species constituted 26.5 and 38.0 per cent of the

TABLE II

The percentage of rats infested, the average number per rat, and the relative percentages of the four most prevalent species of fleas recovered from combined rat samples taken in 17 undusted towns in North Carolina

	DEC.-JAN.-FEB.			MAR.-APRIL-MAY			JUNE-JULY-AUG.			SEPT.-OCT.-NOV.			JAN.-DEC.		
	Rats infest.	Total fleas	Av. no. per rat	Rats infest.	Total fleas	Av. no. per rat	Rats infest.	Total fleas	Av. no. per rat	Rats infest.	Total fleas	Av. no. per rat	Rats infest.	Total fleas	Av. no. per rat
	%	%		%	%		%	%		%	%		%	%	
<i>X. cheopis</i>	29.7	53.7	1.3	28.3	20.6	2.2	43.0	60.9	1.8	47.7	51.9	4.3	34.5	36.1	2.3
<i>N. fasciatus</i>	22.7	26.8	.6	48.4	43.1	3.7	46.9	36.4	1.1	20.1	5.3	.4	33.8	29.1	1.8
<i>L. segnis</i>	9.2	15.6	.4	8.3	9.6	.8	5.5	2.5	.1	6.5	4.6	.3	7.9	8.5	.5
<i>E. gallinacea</i>	2.7	3.7	.1	4.0	26.5	2.2	0	0	0	13.0	38.0	3.1	5.3	26.1	1.6
Number of <i>R. norvegicus</i>	259			324			72			199			854		
Total number of fleas	660			2766			223			1655			5304		

TABLE III

The sequence by per cent (in parentheses) of the three most abundant species of ectoparasites recovered from R. norvegicus in seven undusted towns in North Carolina

	SEASON COLLECTED	NUMBER OF RATS-ECTOPARASITES	FIRST	SECOND	THIRD
Wilson	Fall	45—512	<i>X. cheopis</i> (65.4)	<i>L. bacoti</i> (10.7)	<i>P. spinulosa</i> (8.5)
Wilmington	Fall	37—724	<i>E. gallinacea</i> (27.9)	<i>X. cheopis</i> (25.9)	<i>E. echidninus</i> (21.2)
Clinton	Fall	33—739	<i>A. glasgowi</i> (38.7)	<i>P. spinulosa</i> (34.2)	<i>L. segnis</i> (9.7)
Durham	Fall	55—1163	<i>E. echidninus</i> (49.0)	<i>P. spinulosa</i> (23.0)	<i>X. cheopis</i> (14.0)
Stantonsburg	Spring	33—1683	<i>E. gallinacea</i> (43.3)	<i>P. spinulosa</i> (33.1)	<i>L. segnis</i> (18.2)
Tarboro	Spring	16—723	<i>L. bacoti</i> (35.9)	<i>P. spinulosa</i> (34.3)	<i>X. cheopis</i> (16.4)
Winston-Salem	Spring	36—858	<i>P. spinulosa</i> (66.5)	<i>N. fasciatus</i> (13.4)	<i>X. cheopis</i> (11.0)

total fleas collected in the spring and fall seasons, it infested only 4.0 and 13.0 per cent of the rats trapped during those periods. These data indicate that *E. gallinacea* has a much higher density rate per infested rat than the other species of fleas.

In 11 of the 17 towns, *P. spinulosa* was the most abundant species collected, but in the remaining communities the order of prevalence of the three species having the greatest densities were variable (Table III).

The greatest number of ectoparasites of the various species recovered from *R. norvegicus* was as follows: *P. spinulosa*, 9616; *E. gallinacea*, 543; *N. fasciatus*, 421; *L. bacoti*, 249; *E. echidninus*, 123; *X. cheopis*, 86; and *L. segnis*, 60.

In addition to the species listed from undusted sections, one specimen each of *Pulex irritans* (Linn.), *Hoplopleura hirsuta*, *Hoplopleura oenomydis*, and *Machrocheles* sp. were taken from rats trapped in dusted areas.

EFFECT OF DUST TREATMENTS UPON ECTOPARASITES

In all cases except one, treatment with the 10% DDT dust effectively controlled the oriental rat flea, *X. cheopis*, the common rat flea, *N. fasciatus* (Tables IV and V), and the European mouse flea, *L. segnis* (Table VI). In the one instance where control was not obtained, investigation showed the method of application to be at fault.

Table IV shows that a pre-dust population of 15.3 *X. cheopis* per rat was reduced to 0.3 per rat 11 days after dusting, while 121 days after dusting the average per rat was only 0.1. During the same time, the percentage of rats infested dropped from 57.1 to 11.1 to 7.6. Over a period of 925 days since the initial dusting, nine other applications have been made. In no instance, however, has the *X. cheopis* index been as high as 0.3 per rat. The original pre-dust sample contained 21 rats, from which 323 *X. cheopis* were taken, but the 361 specimens trapped in post-dust samples have yielded only eight *X. cheopis* from four infested rats. Since the oriental rat flea is considered the most important vector of endemic typhus, the excellent control of this species is of much significance.

The 10% DDT dust did not give the same degree of control of the stick-tight hen flea, *E. gallinacea*, as of the rat and mouse fleas (Table VI). The results indicate good control immediately after dusting, but extended residual effect is absent. Observations in other projects have shown similar results.

Eads (1946) treated the roosts, nests, and floors of chicken houses in Texas with the same type dust, and reported complete control of *E. gallinacea* on the chickens within one week. His last observation showed no reinfestation one month after dusting. Gouck (1946) also reported complete control of this species on rats 21 days after treating a poultry store with a similar 10% DDT dust. At the end of 66 days, however, no control was evident, a condition which Gouck attributed to the removal of the fowls which forced the fleas to seek new hosts.

Other species such as the cat flea, *Ctenocephalides felis*, the dog flea, *Ctenocephalides canis*, and *Ctenophthalmus pseudagyrtes* (Baker), were found so infrequently that no data on the effectiveness of the dust in their control could be obtained. It is general knowledge, however, that cat and dog fleas are easily controlled by a 10% DDT dust.

The application of the 10% DDT dust did not successfully control the rat louse, *Polyplox spinulosa* (Table VI). Results similar to those at Stantonsburg were obtained in all communities where dusting programs were in operation. These

data are in line with those of Morlan (1947) who experimented with sulphur and hydroxy pentamethyl flaven added to the 10% DDT dust without obtaining control of this louse.

TABLE IV

The per cent infestation and the average number of X. cheopis per rat recovered from R. norvegicus and R. rattus alexandrinus at Wilson, N. C., before and after premise treatment with 10% DDT dust during the period September 12, 1945, to March 23, 1948

NUMBER OF RATS	DATE DUSTED	AVERAGE NUMBER PER RAT	RATS INFESTED %	DAYS SINCE DUSTED
21		15.3	57.1	0
—	9-14-45	—	—	—
18		.3	11.1	11
14		0	0	40
13		0	0	80
13		.1	7.6	121
—	1-15-46	—	—	—
15		0	0	35
11		0	0	73
8		0	0	98
—	4-26-46	—	—	—
13		0	0	25
9		0	0	53
11		0	0	95
—	8- 2-46	—	—	—
9		0	0	25
—	9-27-46	—	—	—
19		0	0	32
—	11-27-46	—	—	—
15		0	0	13
16		0	0	47
—	1-31-47	—	—	—
18		0	0	26
20		0	0	65
—	4-11-47	—	—	—
13		0	0	41
—	6-15-47	—	—	—
18		0	0	16
24		0	0	51
—	8-12-47	—	—	—
16		0	0	21
16		.1	6.2	91
23		0	0	140
15		0	0	189
14		0	0	223

In all communities, control efforts were directed at those sections which were the foci of typhus cases and which had high rat densities. Because of this practice, check stations established at the periphery of operations did not yield sufficient rats to be of any use in comparative evaluations.

EFFECT OF CONTROL OF ECTOPARASITES UPON SEROLOGY OF RAT

Rats after infection with typhus acquire an immunity which endures for a considerable length of time, and possibly throughout the life of the individual. Philip and Parker (1938) recovered rickettsiae from rat brains 370 days after

TABLE V

The per cent infestation and the average number of X. cheopis and N. fasciatus per rat recovered from R. norvegicus at Dunn, N. C., before and after premise treatment with 10% DDT dust during the period May 1 to July 31, 1946. Dusting date (May 3) indicated by dashes

NUMBER OF RATS	X. CHEOPIS		N. FASCIATUS		DAYS SINCE DUSTED
	Average number per rat	Rats infested	Average number per rat	Rats infested	
29	5.4	48.2	5.4	82.7	0
—	—	—	—	—	—
30	.4	10.0	.1	3.3	12
19	.5	21.1	0	0	34 (20)*
25	.3	12.0	0	0	89 (75)

* Two premises were redusted 14 days after initial dusting.

TABLE VI

The per cent infestation and the average number of L. segnis, E. gallinacea, and P. spinulosa per rat recovered from R. norvegicus at Stantonsburg, N. C., before and after premise treatment with 10% DDT dust during the period April 30 to August 22, 1946. Dust applications (May 1 and August 13) indicated by dashes

NUMBER OF RATS	LEPTOSYLLA SEGNIS		ECHIDNOPHAGA GALLINACEA		POLYPLAX SPINULOSA		DAYS SINCE DUSTED
	Average number per rat	Rats infested	Average number per rat	Rats infested	Average number per rat	Rats infested	
33	5.7	36.3	22.1	33.3	16.9	87.8	0
—	—	—	—	—	—	—	—
19	.2	15.7	3.5	10.5	47.5	73.6	8
14	0	0	46.7	71.3	15.7	100.0	51
12	0	0	51.3	58.3	40.2	75.0	98
—	—	—	—	—	—	—	—
15	0	0	1.6	6.6	4.0	33.0	9

inoculation, while Breinl (1923) found immunity in guinea pigs for a period of 20 months. If the life span of a rat is considered as averaging 3-5 years, it is evident that even with the elimination of transmitting agents, positive blood samples can still be obtained after long periods of time. However, the relative proportion of new-born uninfected rats to mature specimens should gradually cause the percentage of positive seral specimens to drop.

In addition to the time factor, endemicity of the rat population and the non-effectiveness of the dust treatment upon certain types of ectoparasites also

influence the reliability of the results obtained. In certain cities, such as seaports, the influx of new rats into the rat population is probably much higher than in communities which do not have such commercial traffic. While these introduced rats do not have much effect upon the ectoparasitic index unless trapped soon after their arrival, their blood condition can definitely affect the serological results.

As previously mentioned, the 10% DDT dust was much less effective against *P. spinulosa* and *E. gallinacea* than against the rat and mouse fleas. Both of these species are capable of transmitting typhus from rat to rat (Mooser *et al.*, 1931; Brigham, 1941). Another ectoparasite, *Liponyssus bacoti*, has also been shown to transmit and to maintain the disease in rats (Dove and Shelmire, 1931, 1932). Pre-treatment densities of this mite have been low, and subsequent samples immediately after dusting frequently have yielded higher indices. The irregular fluctuations of density indices of this species thus indicate that it is also relatively unaffected by dust treatments. Since *L. bacoti* readily attacks man, these observations are significant.

TABLE VII

The percentage of positive complement fixation tests obtained from samples of rat sera taken at Wilson and Wilmington, N. C., from September, 1945, to March, 1948.

Data grouped in progressive six month periods

	FIRST SIX MONTHS	SECOND SIX MONTHS	THIRD SIX MONTHS	FOURTH SIX MONTHS	FIFTH SIX MONTHS
Wilson.....	33.0 (85)*	14.5 (62)	0 (65)	1.2 (71)	2.4 (78)
Wilmington...	37.5 (72)	19.2 (28)	20.8 (72)	6.2 (48)	22.5 (71)

* Number of rats in sample.

The programs in Wilson and Wilmington are the only ones which have been in continuous operation for the past 2½ years and which initially showed a high percentage of positive rat sera. The data from Wilson (Table VII) indicate a marked reduction 1½ years after the introduction of control measures, and this low rate has been maintained for the past year. The results from Wilmington (Table VII) show a much lesser reduction in percentage during the same period with this decrease being followed by a drop and then a rise in rate for the last 12 months.

In both cities, control of *X. cheopis* has been excellent, and the densities of other species of fleas were not high before or after dusting. No effective control was obtained over either *P. spinulosa* or *L. bacoti*. The rat louse was taken in approximately equal numbers in Wilson and Wilmington, but *L. bacoti* was much more abundant in Wilmington. However, over 95 per cent of *L. bacoti* in Wilmington were taken during the first year of operations.

It does not seem feasible to ascribe the difference in results at Wilmington and Wilson to the lack of control of any species of ectoparasite. It is the writer's belief that Wilmington, a seaport, is more subject to the introduction of outside rats than is Wilson, and that this condition probably has been the most instru-

mental factor in preventing the percentage of positive serological tests from dropping to the low level that has been maintained in Wilson.

SUMMARY

The examination of 854 *R. norvegicus* from 17 towns in North Carolina showed a total of 36,771 ectoparasites, of which the rat louse, *P. spinulosa*, constituted 74.9 per cent. Fleas comprised 14.3 per cent of the total population, with *X. cheopis*, the oriental rat flea, being the most prevalent.

Premise treatments with a 10% DDT dust effectively controlled *X. cheopis*, *N. fasciatus*, and the European mouse flea, *L. segnis*. The stick-tight hen flea, *E. gallinacea*, showed control immediately after dusting, but the residual effects evident in the control of the rat and mouse fleas were lacking. The rat louse, *P. spinulosa*, was not controlled by the dust treatment.

Examination of rat sera for a period of 2½ years at Wilson showed a reduction in the number of positive complement fixation tests from 33 per cent for the first six months to 0-2.4 per cent in the last year and a half. At Wilmington the number of positive tests dropped from 37.5 per cent during the initial six months to 19.2, 20.8, 6.2, and 22.5 per cent respectively for each of the succeeding six-month periods.

LITERATURE CITED

- BENGTSON, I. A. 1941. Complement fixation in endemic typhus fever. Pub. Health Rept. 56(13): 649-653.
- BREINL, F. 1923. Immunity after typhus fever. Med. Klinik 19: 1639-1640. Abst. in Jour. Amer. Med. Assoc. 82: 344, 1924.
- BRIGHAM, G. D. 1941. Two strains of endemic typhus fever isolated from naturally infected chicken fleas (*Echidnophaga gallinacea*). Pub. Health Rept. 56(36): 1803-1804.
- DOVE, W. E., AND B. SHELMIER. 1931. Tropical rat mites, *Liponyssus bacoti* Hirst, vector of endemic typhus. Jour. Amer. Med. Assoc. 97: 1506-1510.
- _____, _____ 1932. Some observations on tropical rat mites and endemic typhus. Jour. Parasit. 18: 159-168.
- EADS, R. B. 1946. Control of the stick-tight flea on chickens. Jour. Econ. Ent. 39(5): 660-661.
- GOUCK, H. K. 1946. DDT to control rat fleas. Jour. Econ. Ent. 39(3): 410-411.
- MOOSER, H., CASTANEDA, M. R., AND H. ZINSSER. 1931. The transmission of the virus of Mexican typhus from rat to rat by *Polyplax spinulosus*. Jour. Exp. Med. 54: 567-575.
- MORLAN, H. B. 1947. Dusts containing combinations of DDT, sulphur, and hydroxy pentamethyl flaven to control rat ectoparasites. Jour. Econ. Ent. 40(6): 917-918.
- PHILIP, C. B., AND R. R. PARKER. 1938. The persistence of the viruses of endemic (murine) typhus, Rocky Mountain spotted fever, and Boutonneuse fever in tissues of experimental animals. Pub. Health Rept. 53(29): 1246-1251.

REACTIONS OF THE ADRENAL CORTEX TO DIPHTHERIA TOXIN

By JOHN NICHOLS

*Departments of Bacteriology and Anatomy, University of North Carolina
Medical School*

Chapel Hill, North Carolina

PLATE 26

Effects of diphtheria toxin on the adrenal glands of experimental animals was first noticed in 1895 by Roux and Yersin, and von Behring (cited from Hoerr, 1931). These changes were described in more detail by DuBois (1896). Since that time many papers have appeared describing the effects of various kinds of stress on the adrenal gland. These have been reviewed by Tepperman (1943) and Selye (1946). Metabolic disturbances during diphtheria toxemia have been reviewed by Holmes (1939).

The purpose of the present investigation is to compare the effects of diphtheria toxin on the lipo-histology of the adrenal gland of a highly susceptible animal and a highly immune animal.

MATERIALS AND METHODS

The results reported here are based on the examination of the adrenals from 30 male guinea pigs and 28 male rats. The guinea pigs were purchased from a dealer and weighed about 250 grams each. Twelve of these animals were injected subcutaneously with 1 ml. of alum precipitated toxoid.¹ All of the guinea pigs were held for one month, by which time immunity had developed in the guinea pigs injected with toxoid. Due to growth since purchase, each of the guinea pigs weighed about 400 grams at this time.

Because the exact MLD concentration of the toxin was not known, 6 normal animals were used in a preliminary titration. They were injected subcutaneously with 1 ml. of the following dilutions of toxin. Two animals received 1 ml. each of a 1-200 dilution, two others 1 ml. of a 1-300 dilution, and the last two 1 ml. of a 1-400 dilution. These animals died 22 hours, 27 hours, and 30 hours respectively after injection. A 1-500 dilution of toxin was prepared and the remaining 24 guinea pigs were injected subcutaneously with 1 ml. During the first 36 hours after injection they were killed as pairs of one immune and one susceptible animal at 6-hour intervals. Forty hours after injection the remaining animals began to die. All guinea pigs were dead within 60 hours.

Twenty-eight male albino rats of the Wistar strain were used, of which 22 weighed 300 grams and 6 weighed 375 grams. None were immunized with toxoid. Fourteen of these rats were injected subcutaneously with 1 ml. of the *undiluted toxin*, the remaining 14 were used as controls. These animals were

¹ Toxin and toxoid kindly furnished by Dr. John H. Hamilton, State Department of Hygiene, Raleigh, North Carolina.

also killed as pairs, one normal animal and one injected animal, at 20-hour intervals until 80 hours after injection, at which time the remaining injected animals began to die. The last animal died 175 hours after injection.

As soon as the animals died or were killed they were immediately autopsied and the adrenals removed and placed in 10 per cent formalin. After remaining in formalin for 24 hours they were removed and the surrounding fat dissected away. One gland from each animal was sectioned with a razor blade, half was embedded in gelatin, sectioned by the freezing method, and mounted according to the technique of Zwemer (1933). These sections were stained with Sudan III according to the method of Romeis (1929) for the demonstration of total lipids. For the demonstration of cholesterol the Schultz reaction was applied to the other frozen sections (Whitehead, 1934a). The other half of the gland was embedded in paraffin, sectioned, and stained with hematoxylin and eosin.

RESULTS

The results of the diphtheria toxin on the adrenal gland as revealed by the lipid stains are described below. The distribution of lipids in the immunized guinea pigs remained normal, as described by Whitehead (1934b). The normal is shown in figure 1. It can be seen that the zona glomerulosa is free from lipids. In the susceptible animals which were injected with toxin and lived longer than 24 hours, numerous fine droplets of lipid may be discerned in the zona glomerulosa (fig. 2). In the animals which lived longer than 50 hours the zona glomerulosa became almost as dense with lipid droplets as the normal zona fasciculata.

The distribution of lipids in the normal rat is shown in figure 3. In the rats which were injected with toxin there occurred a progressive depletion of lipids as the survival time increased. This depletion is more noticeable in the inner zones, while the zona glomerulosa remains relatively refractory. Figure 4 shows the adrenal gland of a rat which lived 140 hours following the injection of toxin.

The distribution of cholesterol in both the guinea pig and the rat as revealed by the Schultz reaction closely resembles the pattern of the total lipids; for this reason they are not shown.

The findings in the hematoxylin and eosin sections were similar to those described by Hoerr (1931). In these preparations the main difference between the guinea pig and rat adrenals is the absence of hemorrhage in the rat adrenal. Glands from both animals show marked hypertrophy, vascular engorgement, and capillary dilation.

DISCUSSION

The best descriptions of the effect of diphtheria toxin on the adrenal gland of the guinea pig are those by Hoerr (1931) and Clevers and Goormaghtigh (1922). These authors did not study the rat.

The writer is unable to explain the reason for the differences in the reaction to the toxin between the guinea pig and the rat. Ingle (1947) points out that the median lethal dose of diphtheria toxin for the rat is 5,000 to 10,000 times

that for the adult guinea pig. Species differences in respect to immunity to various other diseases are also well known.

The appearance of fine droplets of lipid in the zona glomerulosa may be taken as an indication that the gland is reacting to a stress. Knouff *et al* (1941) observed a similar situation in guinea pigs which had been run to exhaustion on a tread mill. Hoerr (1931) who used guinea pigs injected with 1/40 MLD diphtheria toxin observed depletion of lipids after 3 weeks, as did Deansely (1931) who studied the effect of the toxemia of *B. gaertner* (*Salmonella enteritidis*) on white mice. These two latter workers used reduction of osmic acid as an indicator of lipids. In addition to lipids osmic acid is reduced by such substances as glutathione and ascorbic acid. The adrenal gland is a rich storehouse for these two substances (Flexner and Grollman, 1939). Furthermore Sayers (1947) reports that the ascorbic acid content is under the influence of the pituitary.

It is noted that the lipid depletion in the rat is in the inner zones, viz., fasciculata and reticularis. These zones are concerned with the production of the carbohydrate hormone factor of the gland and are under the influence of the pituitary (Swann, 1940). Holmes (1939) points out that there is a marked derangement of carbohydrate metabolism in diphtheria toxemia and that this derangement can, in part, be alleviated by the administration of cortical hormone. He points out that the survival of the animal is enhanced by treatment with cortical hormone. The depletion of lipids in the inner zones may well be connected with the carbohydrate imbalance. Vogt (1947) has shown a similar depletion of lipids in the inner zones of the adrenal cortex of rats by repeated injections of insulin. Most nonspecific stresses such as anoxia cause depletion of lipids in these zones (Nichols, 1948a).

In the guinea pig with the dose of toxin used here the lipid changes consist of an infiltration of small droplets into the glomerular zone. This zone is not under the influence of the pituitary and is concerned with electrolyte balance (Nichols, 1948b; Deane *et al.*, 1948). No explanation can be offered as to why this zone should become infiltrated with lipid droplets in diphtheria toxemia. But as previously mentioned Knouff (1941) observed a similar occurrence in guinea pigs subjected to fatigue.

SUMMARY

Adrenal glands of the highly susceptible guinea pig and highly immune rat have been studied under various degrees of diphtheria toxemia. The lipid changes in the rat consist of a slow gradual depletion in the inner zones of the cortex. The lipid changes in the guinea pig consist of an infiltration of fine droplets into the glomerular zone.

(The writer is greatly indebted to Drs. D. A. MacPherson, F. L. Rights and W. C. George for much valuable advice and criticism and to Miss Charlotte Husted for technical assistance.)

REFERENCES

- CLEVERS, MLE., ET M. GOORMAGHTIGH. 1922. Le cortex surrénal au cours de la vaccination antidiphthérique et de la toxémie diphthérique. Bull. de L'Acad. Royal de Med. de Belgique, Série 5, 2: 425-438.

- DEANE, HELEN WENDLER, JAMES H. SHAW, AND ROY O. GREEP. 1948. The effect of altered sodium or potassium intake on the width and cytochemistry of the zona glomerulosa of the rat's adrenal cortex. *Endocrinology* **43**: 133-153.
- DEANSELY, RUTH. 1931. The histology of adrenal enlargement under experimental conditions. *Amer. J. Anat.* **47**: 475-509.
- DUBOIS, L. A. 1896. Des variations de toxicité des extraits de capsules surrénales. *Arch. de Physiologie Normal et Path.* Cinquième Série Tome Huitième 412-425.
- FLEXNER, LOUIS B., AND ARTHUR GROLLMAN. 1939. The reduction of osmic acid as an indicator of adrenal cortical activity in the rat. *Anat. Rec.* **75**: 207-221.
- HOERR, NORMAND. 1931. The cells of the suprarenal cortex in the guinea-pig. Their reaction to injury and their replacement. *Amer. J. Anat.* **48**: 139-197.
- HOLMES, ERIC. 1939. The effect of toxæmia on metabolism. *Physiol. Rev.* **19**: 439-471.
- INGLE, DWIGHT J. 1947. The resistance of non-adrenalectomized rats to diphtheria toxin with and without adrenal cortical hormone treatment. *Expt. Med. and Surg.* **5**: 375-378.
- KNOUFF, R. A., J. B. BROWN AND B. M. SCHNEIDER. 1941. Correlated chemical and histological studies of the adrenal lipids. The effect of extreme muscular activity on the adrenal lipids of the guinea pig. *Anat. Rec.* **79**: 17-38.
- NICHOLS, JOHN. 1948a. Quantitative histochemical changes in the adrenal following exposure to anoxia. *J. Aviation Med.* **19**: 171-178.
- NICHOLS, JOHN. 1948b. The effect of electrolyte imbalance on the adrenal gland. *Arch. Path.* **45** (June).
- ROMEIS, B. 1929. Weitere untersuchungen zur theorie und technik der sudanfärbung. *Zeitschr. f. Mikr. Anat. Forsch.* **16**: 525-585.
- SAYERS, GEORGE, AND MARION SAYERS. 1947. Regulation of Pituitary Adrenocorticotrophic activity during the response of the rat to acute stress. *Endocrinology* **40**: 265-273.
- SELYE, HANS. 1946. General adaptation syndrome and diseases of adaptation. *J. Clin. Endocrinology* **6**: 117-230.
- SWANN, H. G. 1940. The Pituitary-adrenocortical relationship. *Physiol. Rev.* **20**: 493-521.
- TEPPERMAN, JAY, F. L. ENGLE, AND C. N. H. LONG. 1943. A review of adrenal cortical hypertrophy. *Endocrinology* **32**: 373-402.
- VOGT, MARTHE. 1947. Cortical lipids of the normal and denervated suprarenal gland under conditions of stress. *J. Physiol.* **106**: 394-404.
- WHITEHEAD, RAYMOND. 1934a. The Schultz cholesterol reaction in the suprarenal cortex. *J. Path. and Bact.* **39**: 443-447.
- WHITEHEAD, RAYMOND. 1934b. Variations in the cortical lipid of the guinea-pig suprarenal with sex and age. *J. Anat.* **69**: 72-78.
- ZWEMER, R. L. 1933. A method for studying adrenal and other lipoids by a modified gelatin embedding and mounting technique. *Anat. Rec.* **57**: 41-44.

EXPLANATION OF PLATE 26

All photomicrographs taken of frozen sections stained with Sudan III

Figures 1 & 2 taken at 870 diameters magnification and reduced to 590X by the engraver.

Figures 3 & 4 taken at 135 diameters magnification and reduced to 92X by the engraver.

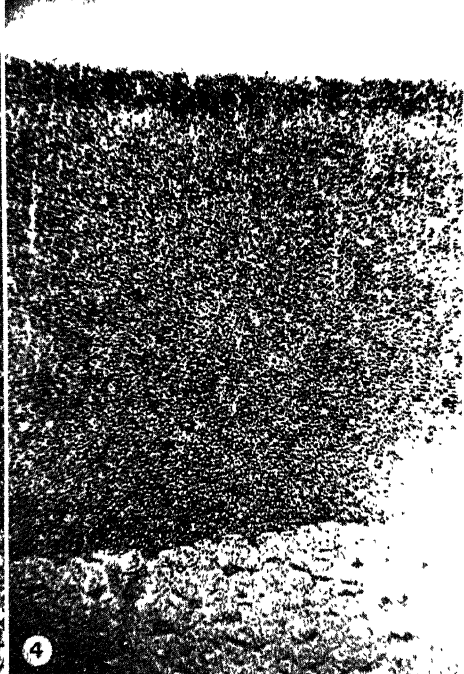
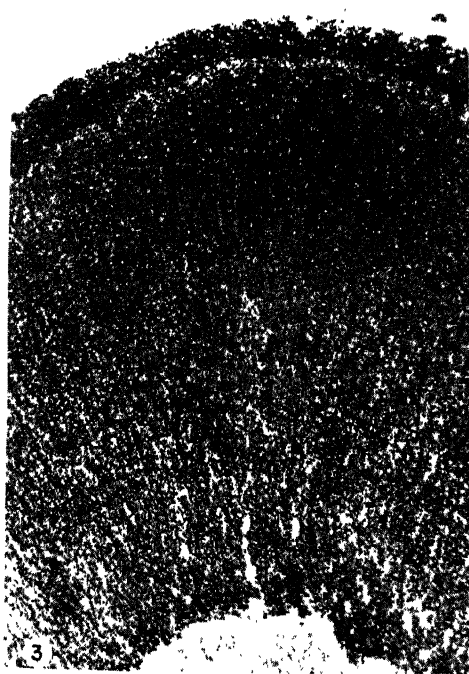
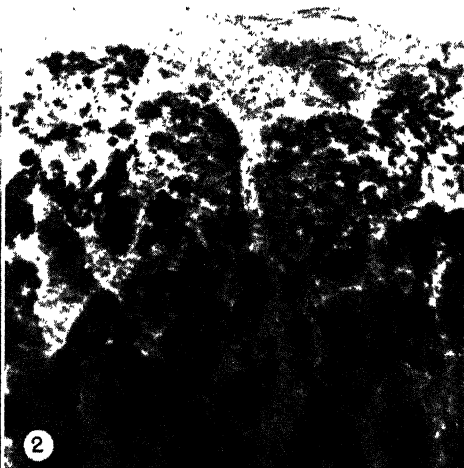
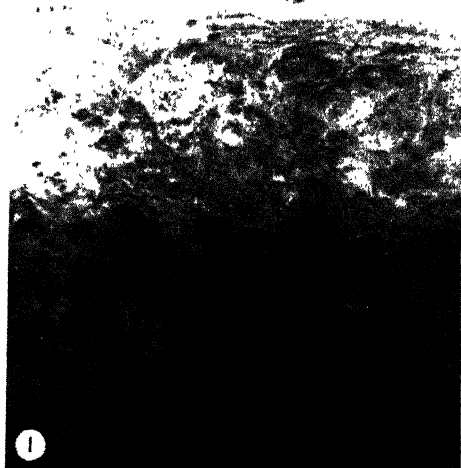
Fig 1. Subcapsular area from adrenal gland of a normal guinea pig. Note absence of lipid in glomerular zone.

Fig. 2. Subcapsular area from adrenal gland of a diphtheritic guinea pig. Note the infiltration of small droplets of lipid into the glomerular zone.

Fig 3. Section through the cortex of a normal rat. Note the normal distribution of lipid.

Fig 4. Section through the cortex of a rat 140 hours after injection of diphtheria toxin. Note the depletion of lipids in the inner zones.

PLATE 26



THE TIGER SHARK, *GALEOCERDO TIGRINUS*, ON THE NORTH
CAROLINA COAST AND ITS FOOD AND FEEDING HABITS
THERE

By E. W. GUDGER

American Museum of Natural History
New York City

FOUR TEXT FIGURES

INTRODUCTION

In an article awaiting publication, I have, among other things, briefly described the Tiger Shark from the standpoint of size and physical characteristics. Suffice it to say here that, as Figure 1 shows, it is grayish in color with dark vertical tiger-like markings. Large specimens run from 10 to 15 feet in total length—15 ft., 2 in. is the largest Western Atlantic specimen and 15 ft., 6 in. is the largest measured tiger shark reported. It is heavily built forward of dorsal fin I, has a large head wider than deep and a very large mouth. This is provided with many rows of highly specialized large serrate sickle-shaped teeth in each jaw, with the points turned outward right and left (Fig. 2). With these teeth, the tiger shark chops up with a shearing bite its sizable prey into large pieces and swallows these through a mouth (distensible horizontally as well as vertically) and a wide gullet into a capacious stomach. I personally know of no other shark that has such a swallowing capacity. This shark is fearless, voracious and omnivorous. It surely merits the colloquial name—"Tiger Shark."

In the article referred to, I described the heterogeneous contents of the stomachs of six tiger sharks, dissected in the summers of 1912-1915, at Key West, Florida. These were mainly caught off Slaughterhouse Point, on the southwest or ocean side of Key West Island. Here the offal from the municipal abattoir attracted numbers of sharks, and particularly the tigers, and these mammalian rejectamenta constituted the major part of the miscellaneous stomach contents. The food of tiger sharks on the North Carolina coast will be found to be very different.

In a third article, in press in the *Australian Museum Magazine*, I have set out the one definite account of the food and feeding habits of *Galeocerdo* in the Tropical Eastern Pacific, and the many records of its feeding in Eastern Australian waters. These accounts extend and confirm the conclusions set out in this and in any other article—that *Galeocerdo* is the most omnivorous and heterogeneous feeder of any shark whose feeding habits have been studied and set forth in a scientific brochure.

Since most of the tiger sharks referred to in this article were taken in the Bight of Cape Lookout, North Carolina, it seems desirable to give Cape Lookout

the setting it deserves as the greatest natural tiger-shark trap in the world. To do this it will be well, as background, to give briefly the few available records



FIG. 1. A 10 ft., 4 in. tiger shark, *Galeocerdo tigrinus*, taken at Tortugas, Florida, May 31, 1913. Note the large head and forepart of the body, the gray side with darker vertical bars (faintly shown). The man beside the shark is 6 ft. tall.

Photograph by Alfred Goldsborough Mayor

of the occurrence and capture of tiger sharks at points in the Western Atlantic north of Cape Lookout; and incidentally to correct a notable error.



FIG. 2. Central part of the lower jaw of a tiger shark 10 ft., 10 in. long, taken at Key West in 1912. Note the large serrate hooked teeth, pointing right and left. These teeth are shown in natural size.

A.M.N.H. photo

THE TIGER SHARK IN THE WESTERN ATLANTIC FROM CAPE COD
TO CAPE LOOKOUT

Galeocerdo tigrinus is indigenous in the tropical Gulf-Caribbean waters. But, in the summers, it migrates in numbers in the warm shallow waters of the Western North Atlantic certainly as far north as the Beaufort-Cape Lookout region of North Carolina. North of this region records are few and brief until, strangely enough, we come to its northern limit. Of it there, Bigelow and Welsh write in their "Fishes of the Gulf of Maine" (1925) that:

Every year a few young tiger sharks are taken in the fish traps in the Woods Hole region, where, according to the records of the Bureau of Fisheries, it is the latest shark to arrive, rarely being seen before August or after October. The specimens captured have usually been about 5 feet long, and very rarely does a full-grown tiger shark stray so far from its tropical home. But, curiously enough, one at least of the several specimens recorded from Provincetown [on the northern extremity of Cape Cod], its farthest known outpost, and the only known locality where it has been captured in the Gulf of Maine, must have been of good size, for its stomach contained a whole grown swordfish.

There seems to be something wrong here. Our authors say elsewhere that adult swordfish run from 7 to 11 feet in length. My largest tiger shark at Key West measured 11 ft., 6 in. between perpendiculars, and the distance along the belly from mouth to vent was 6 ft., 4 in. How much of this was mouth, gullet and stomach (which in *Galeocerdo* form a continuous tube of quite uniform caliber) I do not know—probably it was about half. But, even if this shark had done the improbable and had swallowed such a grown swordfish, then the tail of the fish would have projected from the mouth of the tiger. Indeed, G. P. Whitley, in his "Fishes of Australia, Part I, Sharks," 1940, p. 129, says that a 12-ft. *Galeocerdo rayneri* in Australian waters "had swallowed the 7-ft. tail [only] of a Thresher Shark and part of the tail protruded from the Tiger Shark's mouth when it was caught."

The authors of the "Fishes of the Gulf of Maine" are certainly in error. Such a tiger shark as that referred to by them would probably be of the "20-30 feet length" which some writers allege that *G. tigrinus* attains. Here are the facts back of the error.

In 1864, Capt. N. E. Atwood exhibited before a meeting of the Boston Society of Natural History and presented to it the lower jaw of a large shark captured at Provincetown. This shark, Atwood said, was a *Carcharias*, and because in its stomach had been found "nearly the whole of a fullgrown swordfish," he suggested that it be given the specific name *tigris*. Unfortunately, Atwood did not describe these teeth. And so the matter rested for five years. See Atwood (1864) for the citation.

At a meeting of the Boston Society in January, 1869, Atwood gave a page and a half description of this same *Carcharias tigris*. He also described the teeth, the largest of which "are near the tip of the lower jaw and measured $1\frac{1}{4}$ inches in length, and $\frac{1}{4}$ of an inch in breadth at the base; they are smooth and curve

inwards." With these teeth this shark (8 ft., 10 in. long) in 1864 "had bitten off and swallowed large portions of a swordfish." Here let the reader compare the description of the teeth of *Carcharias tigris*, with Fig. 2 portraying the teeth of *Galeocerdo tigrinus*. The Provincetown fish was tiger-like in his feeding but was not a "tiger shark."

With this introduction, the accounts of the occurrence of tiger sharks on the Atlantic Coast, north of Cape Lookout, will be taken up in order progressing southward. And first, Smith (1898) says that at Woods Hole young ones up to 5 feet long are found in "variable abundance" but "rarely coming before August."

Coming further south our next reference is to Nichols and Murphy (1916) who say that as a straggler *Galeocerdo* is found at Cape Cod, Buzzards Bay, and Vineyard Sound, and note that they have examined the head and jaws of one captured at Islip, on the south shore of Long Island. And more for this study, they record that "The food of the tiger shark is known to include whelks, conchs and squids as well as fishes (menhaden, bonito)."

Of our shark in New York waters Nichols and Breder (1926) repeat the facts of its northern distribution and its food noted above, and add that in New York waters it is "casual in late summer and fall." They give no size of New York specimens.

For New Jersey, Fowler (1907) remarks on its rarity compared with other sharks, and notes that two specimens were once taken at Isle City in early August. Then in 1920 he merely records its occurrence off Cape May and states that he has seen the jaws of two specimens taken off the coast. He gives no data as to sizes attained.

In its northern migration, attested above, *Galeocerdo tigrinus*, must pass through the coastal waters of Virginia and Maryland, or stay further out to sea in the Gulf Stream, but it is significant that in this search no records have been found of its occurrence or capture off the shores of these states.

From the above it seems that, so far as the records go, the tiger shark is so little known north of the North Carolina coast as almost to be considered a foreign fish. Certainly it is a mere straggler. For this paucity of records, and particularly of large specimens, I have an explanation in the possible difficulty that the adult fish, probably rather slow swimmers, would have in finding the relatively large amounts of needed food in the open waters of the Western Atlantic. A better explanation is to be found in the greater abundance of food at Cape Lookout and its ease of capture (both to be emphasized later) which probably serve to detain the large tigers there.

From the North Carolina coast the tiger shark has been recorded, especially from Cape Lookout. But in my day (1902-1911) at the Marine Biological Laboratory of the U. S. Bureau of Fisheries at Beaufort, N. C., it was unknown. The laboratory records did not list *Galeocerdo* in the local fauna and it is not described in Dr. H. M. Smith's "Fishes of North Carolina" (1907). However, the first record of the tiger shark in North Carolina waters is from Beaufort Harbor in

1914. To keep our chronology straight, we must leave the Cape behind for a brief consideration of our shark at Beaufort as recorded in 1916. Incidentally, Beaufort Harbor is below the Cape and about 15 miles away—almost due west (Fig. 3). The presence of *Galeocерdo* there is possibly a reflex of the influence of the Cape as will be seen when we return to a study of that great natural fish trap.

THE TIGER SHARK IN BEAUFORT HARBOR AND ITS FEEDING THERE

In 1916, Lewis Radcliffe made an interesting record of the occurrence, feeding and capture of *G. arcticus* (syn. *tigrinus*) in Beaufort Harbor. In the summer

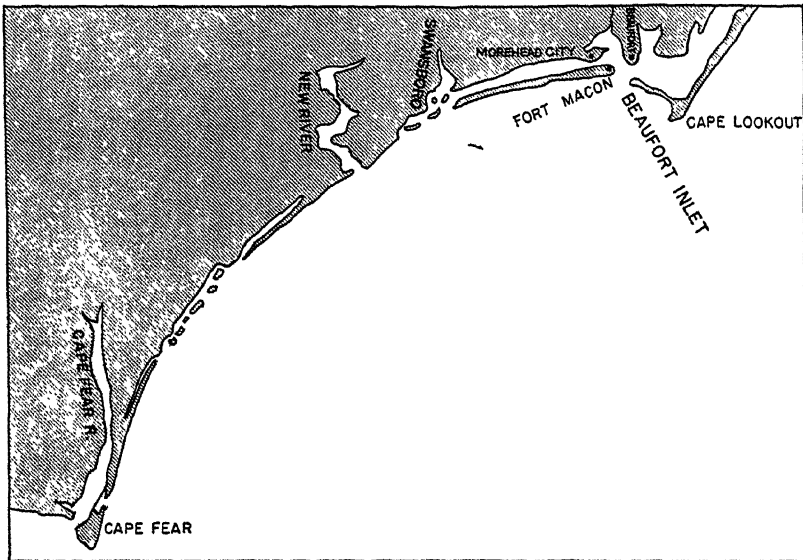


FIG. 3. Outline of the North Carolina coast from Cape Lookout to Cape Fear. The long flat curve (111.5 mi. long) offers no obstruction to tiger sharks moving north in the warm littoral waters. They meet none till they reach Cape Lookout—a natural fish trap.

From U. S. Coast and Geodetic Survey, Diagram No. 8

of 1914 the U. S. Fisheries steamer *Fish Hawk* was anchored off Fort Macon and near the inner mouth of Beaufort Inlet (Fig. 3). Here she was surrounded by a small school of large tiger sharks. They were evidently ravenously hungry, for they swam around the vessel, apparently waiting for anything edible that might be thrown overboard, and they showed no fear of the men moving about but a few feet above them.

A baited shark hook thrown overboard was seized by the largest (and presumably the leader) of the school, who departed "taking bait and hook with him." The next hook was snapped up by another shark, which was brought aboard and killed. The rebaited hook had no sooner reached the water than it was taken by a 10-ft., 1-in. tiger. It was killed and hung up by the tail from the end of the

boom with the head clear of the water. What then took place is vividly described by Radcliffe as follows:

About this time a shark, larger than any of those taken, swam up to the one hanging [by the tail] from the boom, and raising its head partly out of water, seized the dead shark by the throat. As it did so, the captain of the *Fish Hawk*, began shooting at it with a 32-caliber revolver. . . . The shots seemed only to infuriate the shark, and it shook the dead one so viciously as to make it seem doubtful whether the boom would stand the onslaught. Finally it tore a very large section of the unfortunate's belly, tearing out and devouring the whole liver, leaving a gaping hole across the entire width of the whole body large enough to permit a small child to easily enter the body cavity. At this instant, one of the bullets struck a vital spot, and after a lively struggle on the part of the launch's crew, a rope was secured around its tail.

These four tiger sharks, all females, were brought to the laboratory and measured. The largest was 12 ft. and its victim 10 ft., 1 in long. Then, more to our purpose, Radcliffe adds that "at the time of capture, one of the sharks regurgitated a rat, another a small shark about 61 cm. [24 inches] in length."

It has long been known that under certain conditions "shark would eat shark." But, so far as I know, this seems to be the earliest first-hand account of a shark, and particularly a tiger shark, practicing cannibalism on sharks of its own species. These tiger sharks in hunger and fearlessness out-did those which I helped to catch at Key West and there can be little doubt that, had the stomachs of these Beaufort specimens been opened, they would have been found empty. There was no garbage from a large town or city available for them to hunt through, and there was no slaughterhouse near by with a supply of offal. And furthermore in the open ocean, a few hundred yards away, the agile bony fishes had evidently successfully evaded the slower tigers. These big sharks were probably near starvation and were ready to feed on one of their fellows whenever it was held stationary long enough for it to be attacked.

It is interesting to speculate on these questions—"Whence came these sharks?" "How did they chance to come into the relatively shut-off water of Beaufort Harbor?" (See Fig. 3) Radcliffe writes (personal communication) that the *Fish Hawk* had been at anchor (near the inner mouth of Beaufort Inlet) for several days before the tigers appeared. From the anchored vessel all refuse thrown overboard would be carried by the ebb tides out to sea through the Inlet. Then if a band of hungry tiger sharks was working its way north along the coast, when its members scented the waste materials carried out from the ship's galley, they would swim in through the Inlet and surround the ship—as Radcliffe's report states.

I have been unable to ascertain whether other tiger sharks have been taken in Beaufort Harbor since Radcliffe's specimens came through the Inlet in 1914. There is every probability that they have entered the harbor. But if so they do not seem to have been seen, caught, or recorded.

From the above chronological digression, we now return to a study of Cape Lookout and its Bight as a great natural fish trap for tiger sharks migrating

north in the warm, shallow littoral waters of the Western North Atlantic or even in the Gulf Stream. Along with their occurrence, their food and feeding habits will be noted.

CAPE LOOKOUT, NORTH CAROLINA, A NATURAL FISH TRAP

Due to its position and peculiar formation, Cape Lookout and its Bight are functionally a great fish trap. A glance at the map shows that the whole northern half of the coast line of North Carolina makes a great bulge into the Western North Atlantic. From the Virginia-North Carolina line, this barrier-reef sand-bar coast extends in a southeasterly direction to New Inlet and thence almost due south to the much-dreaded Cape Hatteras (Long. $75^{\circ}31'$ W.). From Hatteras, the coast extends in a somewhat southwesterly direction to Cape Lookout (Long. $76^{\circ}32'$ W.), about 70 miles to the west of Hatteras. From Lookout, as Fig. 3 shows, the low sandy coast swings to the southwest in a remarkably great smooth flat curve (111.5 miles long) to Cape Fear. From Cape Fear south to the Florida coast there are no particular prominences to interfere with a northward movement of turtles, sharks, rays, and other fish. Furthermore, from Cape Fear north there is but one inlet (Beaufort) with a saltwater hinterland of size to attract the sharks and other fishes to make a stop off. Attention has already been called to this.

From Fig. 3 it is plain that, after passing Cape Fear, semi-tropical fishes migrating northward in the shallow coastal waters meet with no obstruction till they reach Cape Lookout and its Bight, and the Cape Lookout Shoals extending out to sea. As the chart (Fig. 4) portrays, these form a veritable fish trap into which northbound turtles, sharks, and other fishes naturally herd themselves.

As Fig. 4 shows, the Cape in shape is remarkably like that curious wooden hook used throughout the Central Pacific for catching deep-sea mackerel-like *Ruvettus* fish and large sharks. The base of this hook is V-shaped instead of being U-shaped as in the ordinary hook. From the lighthouse, the straightaway distance (the shank or long leg of the hook) to the base of the Cape Lookout hook is about three miles. The short (or left) leg of the hook, from the pointed base to the barb-region near the bell-buoy, measures about 2.6 miles. And the short leg of the hook and the Lookout Shoals together extend from the bell buoy to the usually travelled north-south steamer lane, a total of about 23 miles—of which the Shoals make up about 20.5 miles. These form a great "leader" to guide northbound fishes into the Bight, which corresponds to the "bag" or "pocket" of a seine, and to the final lethal "room" of a set fish trap (*e.g.*, a pound net). From Fig. 4 and the careful description above, the alleged likeness of the Cape and its Bight (with the long "leader" extending out to sea) to a fish trap is clearly proven. That some tiger sharks may stop off in Beaufort Harbor on the way north to the Cape has been shown. However, most of them persevere till they reach the Bight. And apparently not many large tigers (so far as the records go) seem to cross over or go around the Shoals to migrate farther north on our Atlantic coast.

Furthermore it should be noted that the western edge of the Gulf Stream is distant from Cape Lookout Shoals but 3 or 4 miles. Northward migrating fishes in this great ocean river, feeling the influence of the Shoals, are in many cases likely to swim westward and to follow the "leader" into the great trap of the Bight.¹

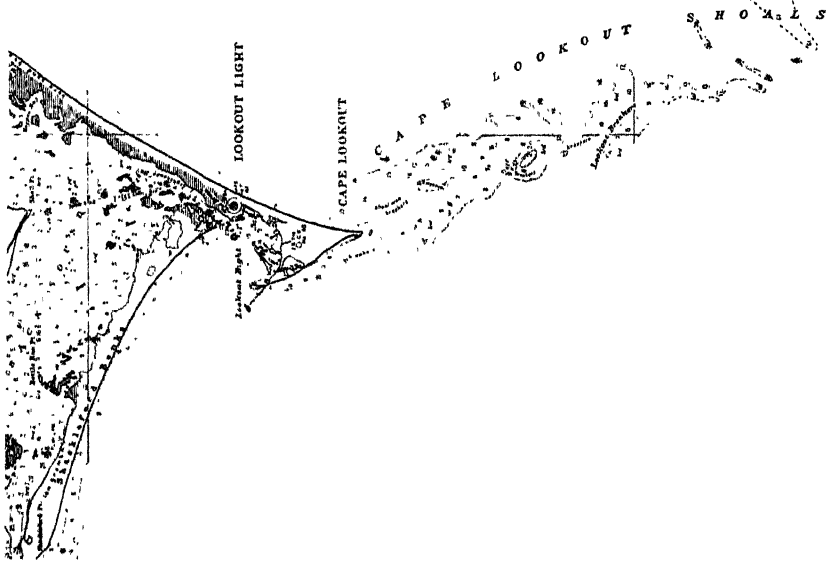


FIG. 4. Cape Lookout, Bight, and Shoals. The Cape has a pointed base instead of a rounded one. The short leg ends in partially enclosing the Bight. The long leg and the Cape Lookout Shoals extend out to sea 20.3 miles (within about 4 miles of the western edge of the Gulf Stream) and form a "leader" to the Bight. Thus all simulate a great pound net or fish trap.

From U. S. Coast and Geodetic Survey, Chart No. 1233

STOMACH CONTENTS OF TIGER SHARKS CAUGHT AT CAPE LOOKOUT, NORTH CAROLINA

The first published account of the tiger shark at Cape Lookout is, so far as this search reveals, from the pen of Russell J. Coles in 1915. As the present writer knows from his long association with the Beaufort Laboratory of the U. S. Fish Commission, Coles was very often fishing at Cape Lookout in the summers from 1902 or 1903 on. Search has been made through all his publications, but not until 1915 is there found a reference to the tiger shark. Here he records one specimen only taken in the latter part of June, 1912. This he identified by teeth and markings, but he did not dissect it for its food. This is the earliest record

¹ For the charts from which Figs. 3 and 4 are copied, and for the mileage measurements noted above, I am indebted to the kindness of Rear Admiral J. H. Hawley, assistant director of the U. S. Coast and Geodetic Survey. For his interest in this paper and for his courtesy in answering many questions, I am very grateful.

(1915) of the occurrence of the tiger shark on the North Carolina coast—and it antedates Radcliffe by two years in discovery (1912–1914) and by one year in publication (1915–1916).

In 1919, Coles made definite record of the food of tiger sharks captured in 1918 in his nets at Cape Lookout. In the stomachs of three young tigers taken May 27, he found pieces of freshly bitten shark meat with adherent skin. In the stomach of the largest (7 ft., 9 in.), there were “eleven of these chunks of shark meat of from one to five lbs. each in weight.” In a large specimen (12 ft., 6 in.) taken July 5, Coles found “parts of three very large stone crabs, a marine bird and other unidentified substances.” Early in August three large tigers (“each in excess of twelve feet in length”) were captured. In one was found “a freshly eaten loggerhead turtle, approximately 100 lbs. in weight, which had been bitten through both shells [carapace and plastron] in three places and the pieces of shell much crushed, yet all of the parts of the turtle were present.”

Herein we have apparently the first definite record of the miscellaneous character of the food of the tiger shark, and the second (Radcliffe, 1916) of the fact that the tiger shark will feed on its fellow sharks.

From 1919–1923, the Ocean Leather Co. operated a shark fishery at Cape Lookout and a shark reduction plant at Morehead City, North Carolina. The sharks were taken at Cape Lookout and presumably mainly in the Bight, but some probably also from around the Shoals. Brought in through Beaufort Inlet to Morehead City, the skins were taken for leather, from the livers was obtained oil for leather dressing, the dried fins went to the Chinese trade, and the flesh was ground up for fertilizer.

And now from this Morehead City plant come the most extensive, carefully made and recorded notes on the food and feeding habits of the tiger shark on the North Carolina coast—or anywhere in the world. These result from the fact that in 1920, Mr. J. C. Bell, of the Department of Preparation in the American Museum, spent several weeks in July and August at Morehead City, the guest of the Ocean Leather Company referred to above. His purpose was to make for the Museum plaster casts of various sharks and rays. But fortunately in addition he recorded lengths (by whom and how accurately measured is not stated) of various sharks captured at Cape Lookout and brought in to Morehead City. He most fortunately also made notes on the stomach contents of many of these sharks and, happily for the purposes of this article, his most abundant notes record the stomach contents of *Galeocerdo tigrinus*.

These notes Mr. Bell and Mr. John T. Nichols, of the Department of Fishes in the American Museum, worked up into a scientific article published in March, 1921, under the title “Notes on the Food of Carolina Sharks.” Later in 1921, seeking a more extensive audience and a wider diffusion of these interesting facts, Mr. Nichols published a general article with the intriguing title—“What Sharks Really Eat.” In this he made use of the data (mainly about the food of the tiger shark) contained in the first article, and from other sources added thereto data for other sharks. Other than my articles on the stomach contents of the tiger sharks in the waters of southern Florida and in the Pacific Ocean, these two papers are the only ones known to me dealing extensively with the food

of *Galeocerdo*. The notes from the article by Bell and Nichols (1921) will now be synopsized—and in their chronological order since this shows the distribution of this shark at Cape Lookout throughout the summer of 1920.

Stomach contents of tiger sharks taken in July, 1920

July 9.—The stomach of a female tiger shark, 11 ft., 3 in. long (weight 636 lbs.) was found to contain the skull of a porpoise, some beef bones and hair, and a large section of the side of the head and gills of an 11-ft. hammerhead taken in the net along with the tiger. July 13.—The stomachs of four tigers contained large pieces of black-tipped sharks bitten in large pieces, some about in half (size not mentioned). July 16.—Two tigers, each about 10 feet long, contained respectively large numbers of horseshoe crab shells, pieces of turtle, three or four small hammerheads, and one of them had part of a net in its stomach. July 17.—Two tigers between 11 and 12 feet long were brought in. One contained partly digested remains of a good-sized shark, and also some pieces of turtle. In the stomach of the other were a piece of turtle, several crabs and some fishes. July 29.—An 11-ft. female had in her stomach several large pieces of bone, much fragmentary turtle shell and the (horny?) jaws of a loggerhead turtle. July 31.—The stomach of a 12-ft., 2-in. female (630 lbs. weight) contained a shark of 8 or 9 feet long bitten in 7 or 8 pieces.

Stomach contents of tiger sharks taken in August, 1920

Aug. 5.—An 11-ft. male (weight 450 lbs.) contained a 3.5-ft. hammerhead intact and parts of several other sharks. A small tiger landed alive on the dock at the same time regurgitated several small mammal bones. Aug. 6.—Two female tigers, 10 ft., 6 in., and 9 ft., 10 in. long, were taken in the same net. The first contained mackerel and a large number of crabs. The other, several small fishes and large pieces of a shark. Aug. 10.—An 11 ft., 7-in. long tiger weighing 553 lbs. contained a large amount of turtle meat and part of a turtle shell, a large number of fresh soft-shelled crabs and various small food fishes. Aug. 12.—Six tigers averaging about 11 ft. long had in their stomachs large sections of porpoises. Contained in the net along with these were six badly mangled porpoises and two intact. Aug. 13.—Three tigers between 10.5 and 11.5 feet long came in. Two contained large pieces of *Mobula* (the smaller two-horned ray). The other had a number of soft-shelled crabs, part of a whip-tailed (sting) ray and several shad. Aug. 16.—Four tigers, averaging about 11 ft., were brought to the dock. One female contained a loggerhead turtle intact (size not noted), another parts of a loggerhead. Several loggerheads were caught in the same net, some badly mangled. Aug. 18.—Seven tiger sharks contained crabs, shad, and many mackerel. One large one had in its stomach a sting ray cut in three pieces, another had bones and feathers of water birds, and the head of the small two-horned ray (*Mobula*). The others were full of small fishes still fresh.

Bell and Nichols (1921) have reported the most complete series of studies ever made on the stomach contents of *Galeocerdo tigrinus*. I shall now quote verbatim the brief synopsis with which they conclude their valuable article.

It will be seen that the above record covers 34 individual Tiger Sharks. Of these thirteen contained crabs, horseshoe crabs, mackerel, shad and other fish, remains of waterfowl (one); two contained mammal bones (probably taken in the role of scavengers); and 26 contained such large food as sea turtles, other sharks, *Mobula* and Whip Ray, and porpoise. Apparently the porpoise, and the other sharks, for the most part, were captured by them in the nets. It is doubtful if they could secure the former or nearly so large a proportion of the latter otherwise.²

That tiger sharks out in the open catch and eat other sharks, seems very doubtful. In none of the tigers which I dissected at Key West, did I find any shark remains, although various other sharks abounded. These specimens were all captured at or near the municipal abattoir from which a steady stream of offal attracted them in numbers to this small area, and supplied them and the other kinds of sharks with an abundant food supply. At the isolated Tortugas the sharks were "on their own," and one incident showed what sharks (in general) would do to one of their fellows when opportunity offered.

One afternoon I put out a well-baited shark hook suspended from a short dry pine log as a float, and made the inboard end of the line fast to a cleat on the stern of our launch. When I visited it next morning I found securely held by the hook—the head of a medium-sized shark. The remainder of the shark was surely entombed in the maws of other sharks.

I think it probable that in all cases where tiger sharks have been found with the remains of other sharks, of rays, and of porpoises in their stomachs, that they and these large marine animals have been ordinarily crowded in comparatively small areas as in the Bight of Cape Lookout, or concentrated in the nets of the fishermen. Here the fierce tigers could readily get at their prey. The same thing applies, of course, to the two cases where hooked sharks were easily attacked by the tigers.

HOW DOES THE TIGER SHARK RID ITSELF OF THE INDIGESTIBLES THAT LOAD ITS STOMACH?

The tiger shark takes everything in through his distensible mouth. In his lower large intestine is a spiral valve which precludes the passage of anything but very soft finely comminuted material. The answer to the question then is that these things must come out through the aperture, the mouth, by which they entered—and by the correlative process of regurgitation. The question is considered at some length in the paper dealing with the Key West tigers, which contained such large indigestible objects as the skulls of a cow and a horse, and the hoofs of horses. However, we find the solution noted twice previously in the present paper. Radcliffe (1916) states that "At the time of capture one of the sharks regurgitated a rat, another a small shark about 61 cm. [24 in.] in length." And Bell and Nichols note that "A small Tiger Shark . . . was landed on the

² In view of the fact that 5 out of 6 of my Key West tigers were females, and that all 4 of Radcliffe's specimens were females, it is much to be regretted that Mr. Bell did not record the sex of each of his specimens. However, in view of his regular work of making casts of the sharks, and his added valuable work of note-taking on sizes and stomach contents, he must have been pressed for time, and this matter did not occur to him.

dock still alive, and vomited up several small mammal bones; leg bones of some sheep have since been identified among them."

The data set out herein definitely establish the fact that the tiger shark, *Galeocerdo tigrinus*, while presumably a migrant only, is a member of the marine fauna of North Carolina. For the present we must consider it only as a summer migrant (in May, June, July, and August) since we unfortunately are lacking records of its occurrence at Cape Lookout for all months other than those of the summer. Possibly and probably it comes earlier and quite surely it stays into September and October.

The data meticulously set out herein definitely establish a new kind of food for the omnivorous tiger shark. This food is conditioned first on the fact that these sharks in their summer migration to the North Carolina coast are more or less concentrated in the Bight of Cape Lookout (a natural fish trap) along with other sharks, rays, porpoises, large marine turtles, and the relatively small bony fishes. Furthermore, all these marine animals are further concentrated when caught in the huge nets of the fishermen. These large nets require time and labor in clearing and while this is being done the fierce tiger sharks are attacking and devouring sharks, rays, porpoises and turtles indiscriminately. These extreme conditions and this feeding by tiger sharks exist nowhere else in the world so far as this search has revealed.

LITERATURE CITED

- ATWOOD, NATHANIEL E. 1864. On the jaws and stomach contents of a large shark, *Carcharias tigris*. Proc. Boston Soc. Nat. Hist. 10: 81.
- ATWOOD, N. E. 1869. Description of a shark, *Carcharias tigris* Atwood. Proc. Boston Soc. Nat. Hist. 12: 268-269.
- BELL, J. C. AND NICHOLS, J. T. 1921. Notes on the food of Carolina sharks. Copeia no. 92: 17-20.
- BIGELOW, HENRY B. AND WELSH, W. W. 1925. Fishes of the Gulf of Maine. Bull. U. S. Fish Comm. 40 (for 1924), part I. (*Galeocerdo arcticus*, pp. 27-28, fig.)
- COLES, RUSSELL J. 1915. Notes on the sharks and rays of Cape Lookout, N. C. Proc. Biol. Soc. Washington 28. (*Galeocerdo tigrinus* at Cape Lookout, p. 89).
- COLES, R. J. 1919. The large sharks of Cape Lookout, North Carolina, etc. Copeia no. 69. (Tiger shark, pp. 38-41, fig. 1).
- FOWLER, HENRY W. 1907. A supplementary account of the fishes of New Jersey. Rept. New Jersey State Museum for 1906. Trenton. (*G. tigrinus*, pp. 255-256).
- FOWLER, HENRY W. 1920. A list of the fishes of New Jersey. Proc. Biol. Soc. Washington 33. (*Galeocerdo arcticus*, p. 144).
- NICHOLS, JOHN T. 1921. What sharks really eat. Natural History 21: 273-278, 7 figs.
- NICHOLS, J. T. AND BREDER, C. M., JR. 1926. The marine fishes of New York and Southern New England. Zoologica (New York) 9. (Tiger shark, pp. 14-15).
- NICHOLS, JOHN T. AND MURPHY, R. C. 1916. Long Island fauna. IV, The sharks. Brooklyn Mus. Sci. Bull. 3. (*G. tigrinus*, pp. 7-8).
- RADCLIFFE, LEWIS. 1916. The sharks and rays of Beaufort, North Carolina. Bull. U. S. Bureau Fish. 34. (Feeding of the Tiger Shark, p. 263).
- SMITH, HUGH M. 1898. The fishes found in the vicinity of Woods Hole. Bull. U. S. Fish Comm. 17. (*Galeocerdo*, p. 88).
- WHITLEY, G. P. 1940. The fishes of Australia, Part I, the Sharks. Sydney. (Tiger shark, p. 113, and other pp.).

A LIST OF CALANOID COPEPODS FROM THE PLANKTON
AT BEAUFORT, N. C.

BY W. H. SUTCLIFFE, JR.

Duke University Marine Laboratory
Beaufort, North Carolina

During the course of a study of the plankton of Beaufort, N. C., the calanoid copepods were identified and recorded. Since the findings of Pearse (1936), little has been reported on the copepods occurring in this locality. This record with notes on seasonal occurrence and relative abundance may help to define the ranges of some of the species. In the interest of further work in this group, a key, adapted from Wilson (1932) and Rose (1933), is given.

The author is indebted to Mrs. M. S. Wilson, Collaborator, U. S. National Museum, for checking some of the identifications and to Dr. C. G. Bookhout of Duke University for valuable criticism.

The collections were made twice monthly from July, 1947, to June, 1948, off Shark Shoal jetty at high and low tides. A Clark-Bumpus plankton sampler with a mesh size of 120 strands to the inch was used.

LIST OF SPECIES¹

Acartia clausii Giesbrecht

Reported by Pearse (1936) as occurring in this region in the months of April and July, 1934. Not found in this investigation.

Acartia longiremus (Lilljeborg)

Reported by Pearse (1936) for April, 1934. Not found in this investigation.

Acartia tonsa Dana

The most common copepod of the plankton at Beaufort. Found from July, 1947, to June, 1948.

Centropages hamatus (Lilljeborg)

Found from December, 1947, to April, 1948.

Centropages typicus Krøyer

Apparently most frequent during the summer months. Observed from July to October, 1947, and May to June, 1948.

Eucalanus monachus Giesbrecht

Occasionally found from July to December, 1947, and in June, 1948.

¹ Except for those reported by Pearse (1936), specimens have been deposited in the U. S. National Museum.

Labidocera aestiva Wheeler

Found in July, 1947, and June, 1948.

Paracalanus parvus (Claus)

Common in the plankton from July, 1947, to February, 1948, and from the latter part of March to June, 1948.

Pontella meadii Wheeler

Found in June, 1948. Rare.

Pseudocalanus minutus (Krøyer)

Reported by Pearse (1936) in the months of May and June, 1934 (as *Pseudocalanus elongatus*). Not found in this investigation.

Pseudodiaptomus coronatus Williams

Found infrequently in July, 1947, and from October to December, 1947.

Temora turbinata (Dana)

Found from July to November, 1947.

Key to the copepods listed

1. Endopods of third and fourth swimming legs of three segments 2
 Endopods of third and fourth swimming legs of two segments 6
2. Endopods of first swimming legs of three segments 3
 Endopods of first swimming legs of two segments 4
 Endopods of first swimming legs of one segment *Pseudocalanus minutus*
3. Terminal exopod segment of third and fourth legs with one lateral spine, one at the distal corner and one terminal *Pseudodiaptomus coronatus*
 Terminal exopod segment of third and fourth legs with two lateral spines, one at the distal corner and one terminal *Centropages* 9
4. Caudal rami at least six times as long as wide; fifth legs uniramous and of three segments in the female; in the male left fifth leg of four segments, chelate, right of three segments *Temora turbinata*
 Caudal rami only three times as long as wide or less 5
5. Middle segments of third and fourth endopods with two, end segments with seven, setae *Paracalanus parvus*
 Middle segments of third and fourth endopods with one, end segments with five, setae *Eucalanus monachus*
6. Endopod of first legs made up of three segments *Pontella meadii*
 Endopod of first legs made up of two segments 7
7. Head with one pair of dorsal lenses; fifth legs biramous in the female, rami of one segment and unequal; right leg uniramous in male, with chela *Labidocera aestiva*
 No dorsal lenses; fifth legs not segmented as above 8
8. Fifth legs biramous in both sexes *Centropages* 9
 Fifth legs uniramous in both sexes 10
9. Segments one, two, and five of first antennae with a spine *Centropages typicus*
 These segments without spines *Centropages hamatus*
10. Caudal rami at least six times as long as wide; fifth legs of three segments in female; in male left fifth leg of four segments, right of three segments *Temora turbinata*
 Caudal rami only three times as long as wide or less *Acartia* 11

NOTES ON THE DISTRIBUTION OF *CARYA AQUATICA* IN NORTH CAROLINA

BY WILLIAM B. FOX

*Department of Botany, North Carolina State College
Raleigh, North Carolina*

PLATES 27, 28, AND ONE TEXT FIGURE

Carya aquatica (Michx. f.) Nuttall seems to have been as little collected in North Carolina as any tree whose range is known to cross a portion of that state. According to Sargent in his *Silva of North America*, it has been known since 1875 to occur as far north in the Atlantic Coastal Plain as Mobjack Bay in Virginia. In Sargent's *Manual of the Trees of North America* the range is given as in coastal regions from Virginia to eastern Texas (Brazos River), and northward in the Mississippi Valley to southeastern Missouri and Southern Illinois.

During the summer of 1947, in the course of making a collection of woody plants in North Carolina, I became interested in locating trees of this species. Examination of the herbaria of the University of North Carolina, Duke University, and North Carolina State College yielded specimens from only three localities in the state: one from the Cape Fear River Valley in Cumberland County and two from the vicinity of Wilmington near the same river. Coker and Totten in their *Trees of the Southeastern States* report it as being "known from the Cape Fear and Pamlico Sound regions."

The larger herbaria of eastern United States have a very poor representation of *Carya aquatica* from North Carolina. By personal examination or by correspondence with curators of the institutions, I learned the following facts, as of the fall of 1947. There were no specimens from this state in the herbarium of the New York Botanical Garden nor in that of the Chicago Museum of Natural History. The herbarium of the Arnold Arboretum had one: a collection by T. G. Harbison (No. 12) from the "Neuse River Swamp, Goldsboro, North Carolina". The U. S. National Herbarium and the Gray Herbarium had one each, viz., R. K. Godfrey and B. W. Wells #4890, from Brunswick County, near Wilmington, collected July 9, 1938.

The habitat of *Carya aquatica* is given in most manuals as "deep swamps" or "river swamps". Coker and Totten (1945) give "deeper swamps of the coastal plain"; Sargent's Manual (1933) has it "river swamps often inundated a considerable portion of the year"; Gray's Manual (7th ed.), "river swamps", and Small's Manual (1933) lists "river swamps, river bottoms and moist or wet woods". Other manuals describe the habitat in similar terms.

I have had an opportunity, with the help of B. W. Wells and L. A. Whitford, to explore by canoe considerable portions of what we consider to be deep, river swamps in the North Carolina coastal plain, e.g., Lockwood Folly River and Town Creek in Brunswick County. We now have collections from 16 counties in North Carolina, including most of the major streams of the state, and none

of them are from a habitat that could accurately be described as a "deep swamp". I have not seen it growing in close proximity with such typically swamp species as *Fraxinus caroliniana* Mill., *F. tomentosa* Michx. f., and *Nyssa aquatica* L., nor have I seen it standing in water as is often the case with these species. These swamp trees in some places were in the general vicinity, but always in lower and much wetter situations.

In all locations where the tree was found, it was on banks a few feet above the water level, on ground that would be covered by water only during flood stages of the stream (Plate 27). However, it was not found far from streams in any case nor usually more than approximately eight or ten feet above the summer water level. Flood plain is a more appropriate term for its usual habitat in the localities where I have seen the tree. Common associates of *Carya aquatica* in the coastal plain of North Carolina are *Carpinus caroliniana* Walt., *Ilex decidua* Walt., *Acer Negundo* L., *Platanus occidentalis* L., *Quercus lyrata* Walt., *Celtis laevigata* Willd., and *Carya cordiformis* (Wang.) K. Koch.

This hickory was usually quite plentiful in the localities where we observed it, in some places along the Neuse River being one of the most common river-bank trees. The largest specimen seen is one at Richardson's Bridge on the Neuse River in Johnston County, west of Goldsboro. It measured 38 inches in diameter at breast height (Plate 28).

We have been able to determine what is probably the farthest upstream that the tree grows on the Neuse River and the Tar River by traversing a considerable length of these rivers by canoe. After one becomes accustomed to the aspect of this hickory, it can be recognized at a considerable distance. I have not found it on the Neuse River above a point approximately two miles down-river from Smithfield in Johnston County. Below this point it was quite plentiful at every location visited between there and New Bern, in Craven County—seven places in all.

On an estimated thirty-mile canoe trip on the Tar River this past summer between a point below Rocky Mount and the N. C. Rt. 44 bridge north of Tarboro, L. A. Whitford and I failed to find *Carya aquatica* until we had reached a point about five miles above the bridge just mentioned. We failed to find it on a similar trip of at least fifteen miles on the Cape Fear River in Cumberland County from a point west of Wade to Fayetteville. The Cape Fear in this region is fairly deeply entrenched with precipitous banks, in some places twenty-five to thirty feet above the water level. One may safely say that *Carya aquatica* does not occur on the Cape Fear River above Fayetteville, though Dr. H. R. Totten of the University of North Carolina has collected it on Rockfish Creek near its junction with this river at a point approximately 8 miles below Fayetteville.

A most interesting locality for this hickory was found this past summer by Steve G. Boyce, a forestry student at North Carolina State College. He found several trees on the west side of the Yadkin (or Pee Dee) River at a point about two miles east of N. C. Rt. 109. Though still in the coastal plain, this is considerably farther inland than any other locality known for this species in North

Carolina. These trees were growing on higher ground at the margin of an old drainage ditch about one thousand feet from the river.

Since details of the distribution of *Carya aquatica* in North Carolina seem previously to have been virtually unknown, or at least undocumented by herbarium specimens, a map showing the counties in which we have collected it, is herewith published (Fig. 1). Following is a list of all the collections from this state of which I am at present aware.

WASHINGTON Co.: sandy beach, Albemarle Sound, near s. end of N. C. Rt. 32 bridge, May 21, 1948, *W. B. Fox* and *R. K. Godfrey* #1547 (a single depauperate specimen). MARTIN Co.: alluvial plain, bank of Roanoke R., n.e. of Hamilton, May 21, 1948, *W. B. Fox* and *R. K. Godfrey* #1498. HALIFAX Co.: higher ground between gum swamp and creek, Fishing Creek, 2 mi. w. of U. S. Rt. 258, May 22, 1948, *W. B. Fox* and *R. K. Godfrey* #1630. EDGECOMBE Co.: low,

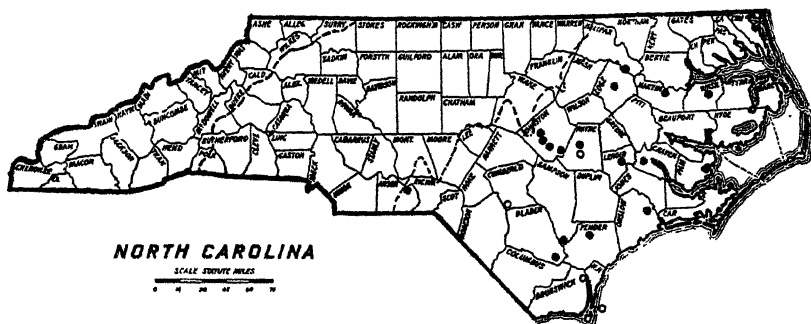


FIG. 1. Map of North Carolina showing distribution of collections; open circles indicate collections previous to summer of 1947, solid circles since summer of 1947.

sandy bank, Tar R., 5 mi. above N. C. Rt. 44 bridge, June 17, 1948, *W. B. Fox* and *L. A. Whitford* #1748. CRAVEN Co.: higher sandy bank of Neuse R., at mouth of Batchelder Cr., Aug. 31, 1947, *W. B. Fox* and *L. A. Whitford* #1123. LENOIR Co.: sandy bank of Neuse R., just below U. S. Rt. 70 br., s. of Kinston, Aug. 31, 1947, *W. B. Fox* #1141. WAYNE Co.: sandy bank of Little R., just above Rt. 70, Aug. 31, 1947, *W. B. Fox* #1144; Neuse River Swamp, Goldsboro, *T. G. Harbison* #12. JOHNSTON Co.: near Richardson's Bridge, Neuse R., Oct. 4, 1947, *W. B. Fox*, *L. A. Whitford* and *R. J. Campana* #1239; sandy bank of Neuse R., Boone Twp., 9 mi. below Smithfield, Nov. 1, 1947, *W. B. Fox*, *R. K. Godfrey* and *L. A. Whitford* #1273; overhanging Neuse R., below U. S. Rt. 301 br., Nov. 1, 1947, *W. B. Fox*, *R. K. Godfrey* and *L. A. Whitford* #1287; low bank of Neuse R., about 2 mi. below Smithfield, Nov. 8, 1947, *W. B. Fox* and *Bert Whitford* #1302. ONSLOW Co.: bank of Whiteoak R., e. of Belgrade, June 21, 1948, *L. A. Whitford*. PENDER Co.: flood plain, N. E. Cape Fear R., near bridge s. e. of Angola Bay, June 10, 1948, *W. B. Fox*, et al, #1728. NEW HANOVER Co.: near Cape Fear R., Carolina Beach, July 7, 1940, *B. W. Wells*. BRUNSWICK Co.: river bank, near Wilmington, July 9, 1938, *R. K. Godfrey* and *B. W. Wells* #4890. BLADEN Co.: n. bank of Cape Fear R., $\frac{1}{2}$ mi. above ferry, n. e.

of Carvers, Aug. 28, 1948, *W. B. Fox* and *L. A. Whitford* #1857. SAMPSON Co.: in edge of road fill, Newkirk's Br., Black River, n. of Ivanhoe, Aug. 29, 1948, *W. B. Fox* and *L. A. Whitford* #1905. CUMBERLAND Co.: bank of Rockfish Cr. near mouth, June 28, 1937, *H. R. Totten*, et al. ANSON Co.: higher ground at margin of old drainage ditch near Yadkin (Pee Dee) River, 2 mi. s. of N. C. Rt. 109, July 27, 1948, *Steve G. Boyce* #1022.

Carya aquatica thus is much more widely distributed in North Carolina than collections previous to 1947 would indicate. The collections listed here and our observations as to its abundance in many localities show that it is one of the common river-bank trees in the coastal plain of this state. My experience with the tree lead me to suggest that the habitat designation of "deep swamp" and "river swamp" given for this hickory in most manuals, is misleading. At least, where I have seen it in North Carolina, *Carya aquatica* is most commonly found on the stream-side margins of the flood plains of the coastal plain rivers.

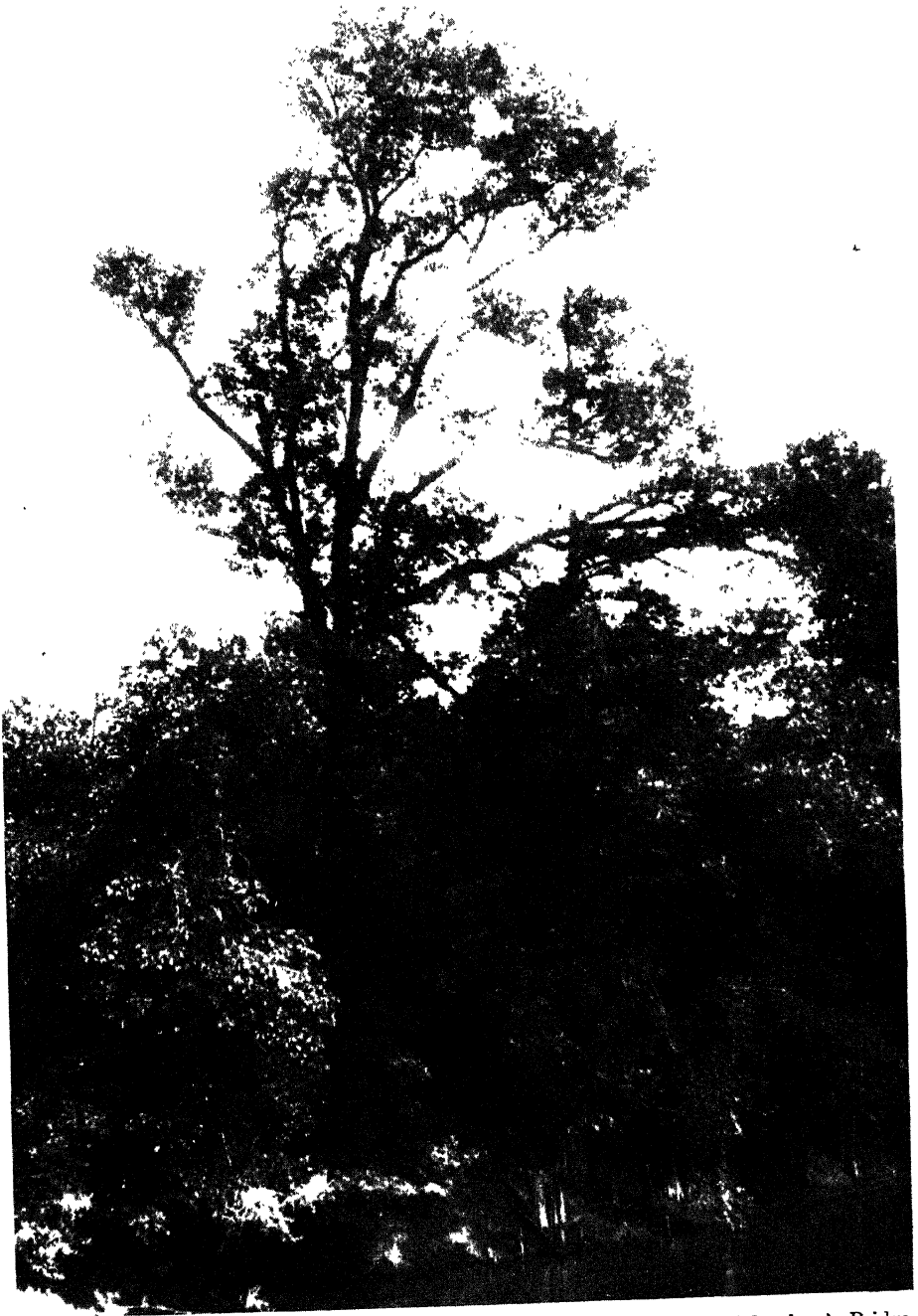
ACKNOWLEDGMENTS

I am indebted to Dr. L. A. Whitford for assistance in preparing the figure and plate and to Dr. B. W. Wells for helpful suggestions.

LITERATURE CITED

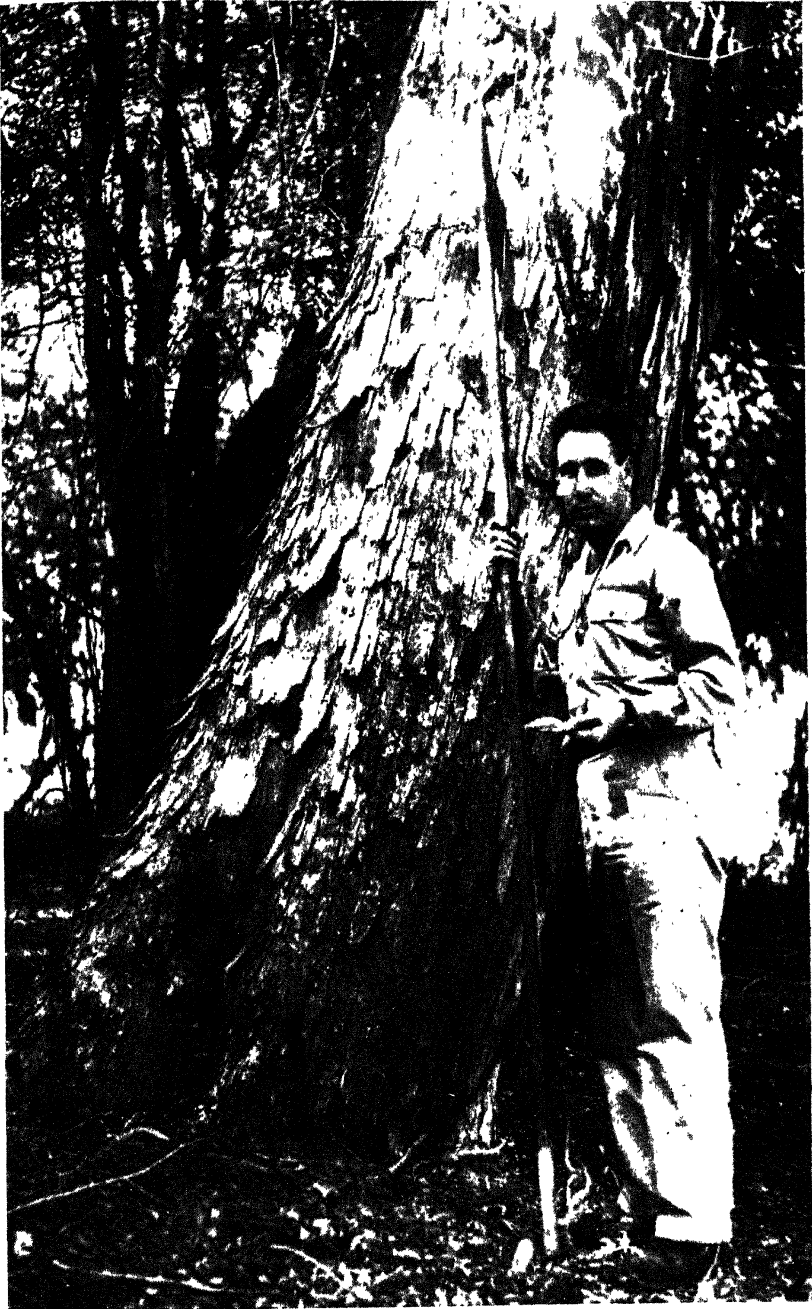
- COKER, W. C., AND H. R. TOTTON. 1945. *Trees of the Southeastern States*, 3rd Edition, Univ. N. C. Press, Chapel Hill.
- ROBINSON, B. L., AND M. L. FERNALD. 1908. *Gray's New Manual of Botany*, 7th Edition. American Book Co., New York, N. Y.
- SARGENT, C. S. 1933. *Manual of the Trees of North America*. Houghton Mifflin & Co., New York, N. Y.
- SARGENT, C. S. 1895. *The Silva of North America*, vol. VII. Houghton Mifflin & Co., New York, N. Y.
- SMALL, J. K. 1933. *Manual of the Southeastern Flora*. Published by the Author, New York, N. Y.

PLATE 27



A medium-sized tree of *Carya aquatica* on bank of Neuse River, near Richardson's Bridge, Johnston County, N. C., showing typical habitat.

PLATE 28



Trunk of large specimen of *Carya aquatica*, same location as other one shown. This trunk measured 38 inches in diameter at breast height.

STUDIES IN THE COMPOSITAE OF NORTH CAROLINA

I. *Liatris*

BY R. K. GODFREY

*Department of Botany, North Carolina State College
Raleigh, North Carolina*

INTRODUCTION

The following brief treatment of *Liatris* in North Carolina is based largely upon the recent monograph of Gaiser.¹ This excellent work with its painstaking citation of documenting herbarium specimens offers an opportunity for separating out pertinent information concerning the local taxonomic entities and their known distribution in the state. This has been done here and incorporated with it are additional data obtained by me when, using the monograph, I examined the specimens in the herbaria of the North Carolina State College, the University of North Carolina, and Duke University, many of which, apparently, Dr. Gaiser did not see.

An artificial key to the North Carolina species and varieties of *Liatris* is presented below. Following that the distributional data are summarized on outline maps of the state, Dr. Gaiser's citations being indicated by solid dots, and the additional specimens which I have seen and interpreted being indicated by open circles. On these maps a statement of general distribution is also given. It is hoped that the key together with the summaries charted on the maps will be helpful to field workers. Synonymy is given only where names are quite unlike those under *Laciniaria* in Small's Manual (1933). It is not felt that detailed descriptions should be included here. A few observations made in the field are added because it is thought they present facts not previously recorded or at variance with Dr. Gaiser's conclusions.

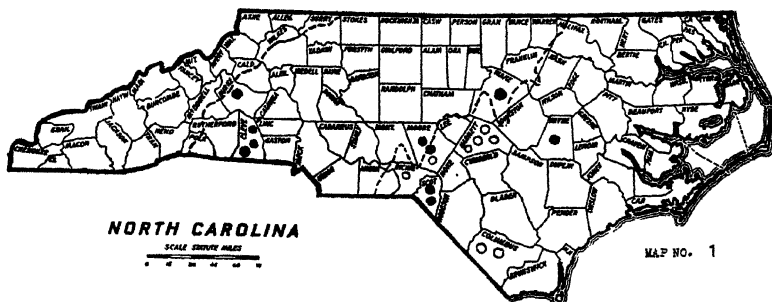
Key to Liatris in North Carolina

- a. Pappus barbellate, the lateral cilia only a few times longer than the diameter of the setae.
- b. Heads with 3-20 flowers, turbinate-campanulate, or oblong-cylindrical, not over 1 cm. broad.
 - c. Inner phyllaries (involucral bracts) acuminate.
 - d. Heads with 6-12 flowers, not over 1 cm. long, not secund, sessile or short-peduncled; outer phyllaries keeled, pappus 3-4 mm. long
 - 1. *L. regimontis*, Small
 - d. Heads with 3-6 flowers, up to 1.5 cm. long, often secund, on peduncles 1-3 cm. long; outer phyllaries not keeled; pappus 8-9 mm. long ... 2. *L. secunda*, Ell.
 - c. Inner phyllaries rounded at apex or obtuse.
 - e. Inner phyllaries broadly rounded at the apex.
 - f. Pappus only half the length of the corolla tube, not showing beyond the involucre at the time of flowering 3. *L. Helleri*, Porter
 - f. Pappus more than half the length of the corolla tube, exerted from the involucre at the time of flowering.
 - g. Heads oblong-cylindrical; inflorescence spicate, or very rarely with the lowermost heads short-peduncled.

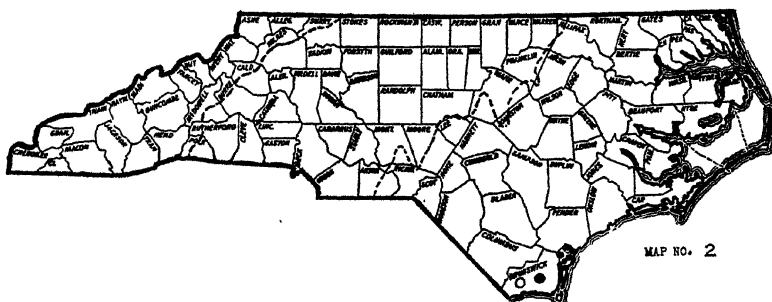
¹ Gaiser, L. O., The Genus *Liatris*, *Rhodora* 48: August-December 1946.

- h. Lower cauline leaves long-lanceolate; heads with 10-18 flowers; longer involucre 1 cm. or more long; phyllaries green or only slightly purplish.
- i. Spikes elongate up to 2-7 dm. long; lower cauline leaves little more than a dm. long. . . 4. *L. spicata* (L.) Willd., var. *typica* Gaiser
- i. Spikes short and thick; characteristically about 3 dm. long; lower cauline leaves 2-4 dm. long.
- 5. *L. spicata* (L.) Willd., var. *typica* Gaiser, forma *montana* (Gray) Gaiser.
- h. Lower cauline leaves linear; heads with 4-7 flowers; longer involucre not over 7 mm. long; phyllaries usually strikingly purplish.
- 6. *L. spicata* (L.) Willd., var. *resinosa* (Nutt.) Gaiser.
- g. Heads turbinate; inflorescence spicate, racemose, or paniculate.
- j. Inflorescence an elongate, many-headed raceme or panicle; leaves gradually diminishing in size up the stem.
- k. Leaves linear, rarely linear-lanceolate; stem and leaves with scattered hairs; heads up to 8 mm. broad; phyllaries 1 mm. broad.
- 7. *L. graminifolia* (Walt.) Willd., var. *typica* Gaiser.
- k. Leaves linear-lanceolate; stem and leaves glabrous except for long hairs on the petioles; heads 1 cm. broad; phyllaries 2 mm. broad.
- 8. *L. graminifolia* (Walt.) Willd., var. *dubia* (Barton) Gray
- j. Inflorescence a short virgate spike with a few scattered, sessile heads; leaves abruptly reduced below the spike
- 9. *L. graminifolia* (Walt.) Willd., var. *Smallii* (Britton) Fern & Grisc.
- e. Inner phyllaries obtuse.
- l. Lower cauline leaves narrowly linear; heads numerous in lax, slender racemes, the peduncles about equalling the heads in length; heads narrowly oblong or cylindrical; 4-6-flowered; pappus about half the length of the corolla tube.
- 10. *L. microcephala* (Small) K. Sch.
- l. Lower cauline leaves linear-lanceolate; heads distant, sessile or peduncled, forming a stiff open spike or raceme; heads turbinate, 9-20-flowered; pappus nearly as long as the corolla tube 11. *L. turgida* Gaiser
- b. Heads with 15-50 flowers, broadly turbinate to campanulate-hemispheric, over 1-5 cm. broad.
- m. Outer phyllaries ovate, obtuse; inner phyllaries oblong-spatulate, rounded; all herbaceous. 12. *L. Earlei* (Greene) K. Sch.
- m. Outer phyllaries orbicular, oblong, or ovate, rounded; inner phyllaries oblong-spatulate to obovate; herbaceous or with scarious margins.
- n. Leaves linear-lanceolate to lanceolate, glabrous or with few scattered hairs; stem glabrous below, with appressed hairs on rachis or inflorescence.
- 13. *L. aspera* Michx., var. *intermedia* (Lunnell) Gaiser
- n. Leaves lanceolate to oblanceolate, often oblong to obovoid, sparingly to densely pubescent; stem with dense semi-appressed pubescence throughout.
- o. Heads broadly turbinate, 15-25-flowered; outer phyllaries markedly squarrose; pappus 5-6 mm. long.
- 14. *L. scariosa* (L.) Willd., var. *squarulosa* (Michx.) Gray
- o. Heads campanulate-hemispheric, or subglobose, 20-30-flowered; outer phyllaries slightly if at all squarrose; pappus 6-9 mm. long.
- 15. *L. scariosa* (L.) Willd., var. *virginiana* (Lunnell) Gaiser
- a. Pappus plumose, the lateral cilia many times the diameter of the setae.
- p. Heads thick and almost isodiametric in outline; outer phyllaries markedly squarrose.
- 16. *L. squarrosa* (L.) Michx.
- p. Heads cylindrical; outer phyllaries not squarrose.
- 17. *L. squarrosa* (L.) Michx., var. *gracilentia* Gaiser

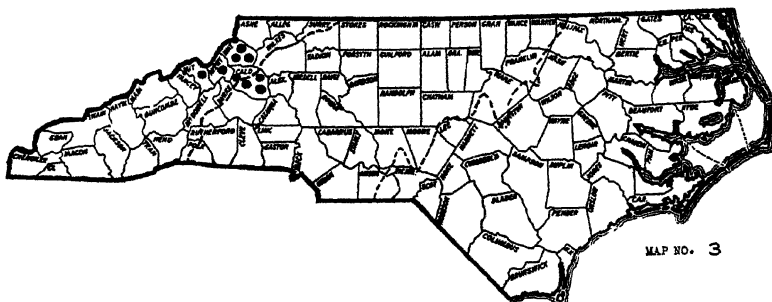
MAPS SHOWING DISTRIBUTION BY COUNTIES OF THE SPECIES AND VARIETIES OF
LIATRIS IN NORTH CAROLINA



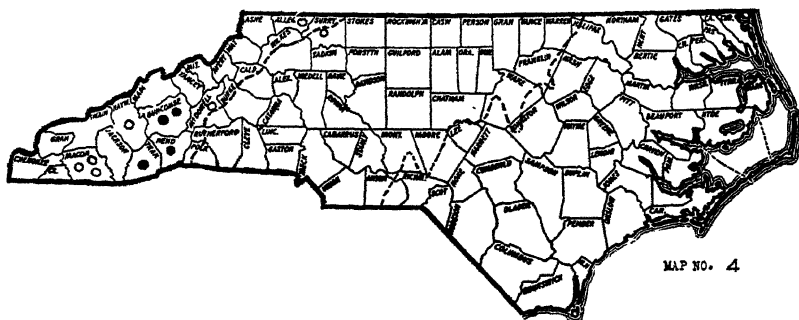
MAP No. 1. *Liatris regimontis* (Small) K. Sch. Eastward from the mountains in Virginia, North Carolina, and South Carolina.



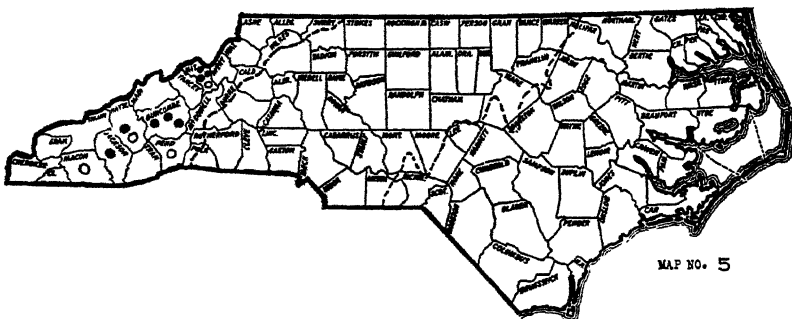
MAP No. 2. *Liatris secunda* Ell. From the coastal plain region in North Carolina through South Carolina, Georgia, and central and eastern Florida to Alabama and Louisiana.



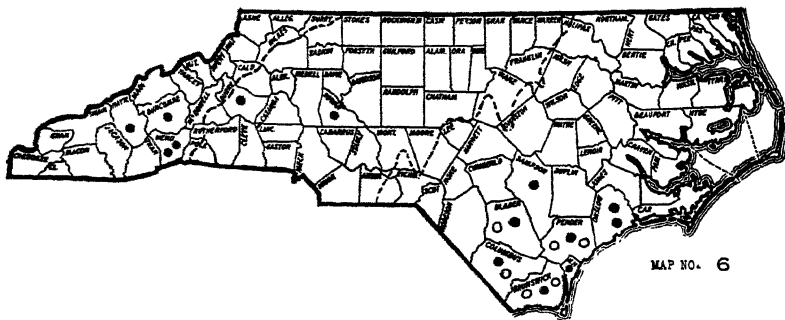
MAP No. 3. *Liatris Helleri* Porter. Only in the mountains of North Carolina.



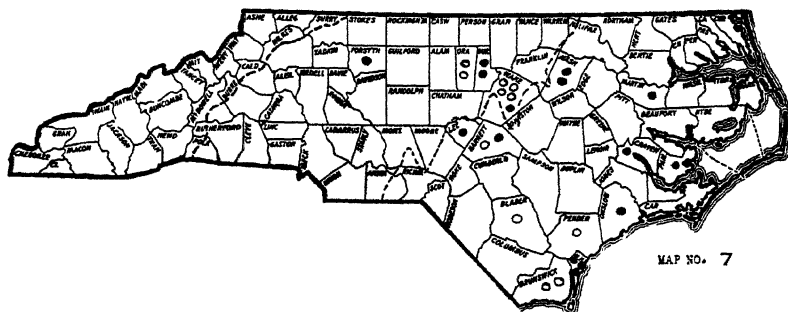
MAP No. 4. *Liatris spicata* (L.) Willd., var. *typica* Gaiser. From Long Island, New York, to Florida, and westward to the Mississippi; from the Lake St. Clair region of Ontario and Michigan to the Gulf of Mexico.



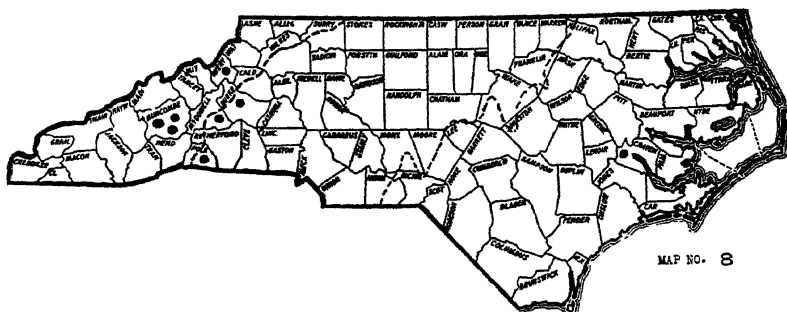
MAP No. 5. *Liatris spicata* (L.) Willd., var. *typica* Gaiser, forma *montana* (Gray) Gaiser. Mostly in the mountains of North Carolina.



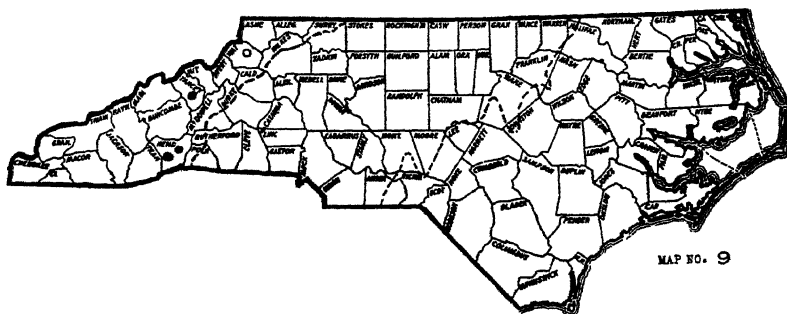
MAP No. 6. *Liatris spicata* (L.) Willd., var. *resinosa* (Nutt.) Gaiser. Pennsylvania to Tennessee, south to Florida and Mississippi.



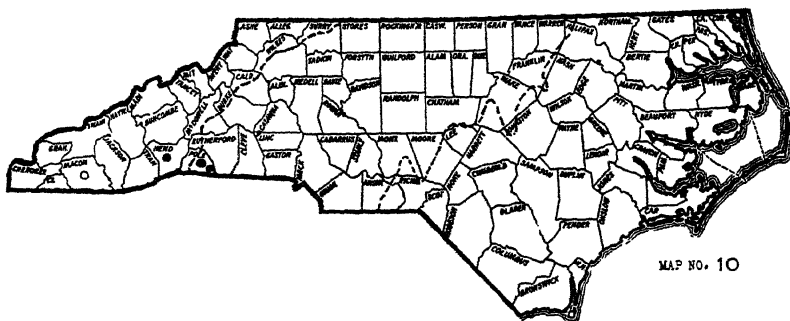
MAP No. 7. *Liatris graminifolia* (Walt.) Willd., var. *typica* Gaiser. New Jersey to Alabama, chiefly on the coastal plain.



MAP No. 8. *Liatris graminifolia* (Walt.) Willd., var. *dubia* (Barton) Gray. New Jersey to Pennsylvania, southward through North Carolina.

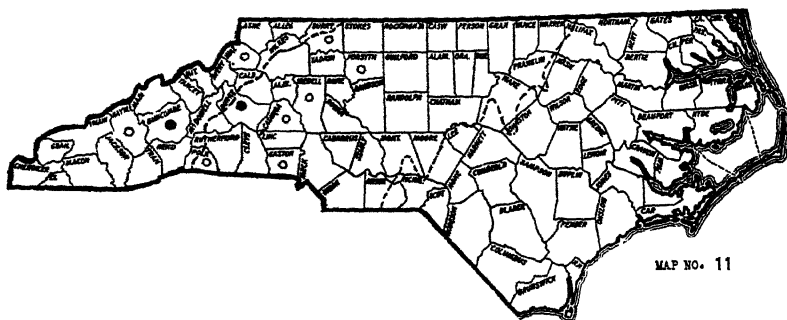


MAP No. 9. *Liatris graminifolia* (Walt.) Willd., var. *Smallii* (Britton) Fern and Griseb. (*Laciniaria Smallii* Britton) Southwestern Virginia to Georgia, chiefly in the mountains.



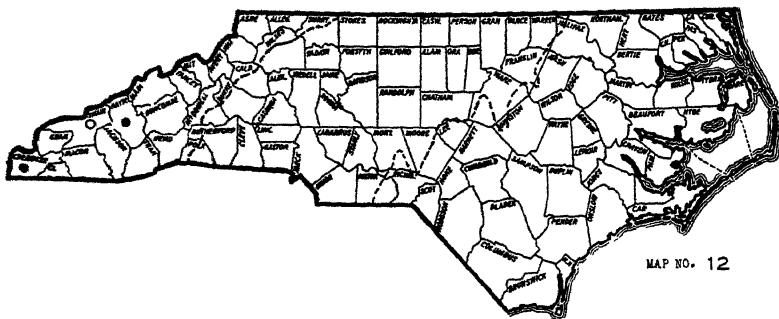
MAP NO. 10

MAP No. 10. *Liatris microcephala* (Small) K. Sch. On sandstone outcrops and dry barrens in North Carolina, Tennessee, Kentucky, Georgia, and Alabama.



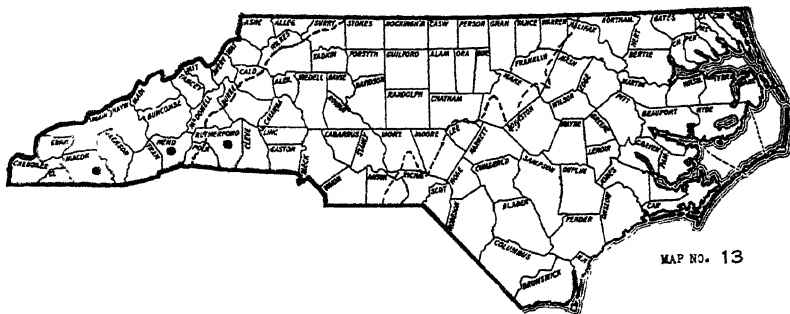
MAP NO. 11

MAP No. 11. *Liatris turgida* Gaiser. Mountain ridges of southwestern Virginia and adjoining West Virginia and North Carolina.



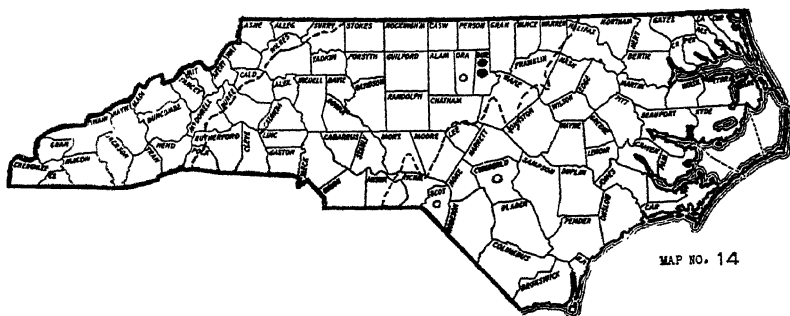
MAP NO. 12

MAP No. 12. *Liatris Earlei* (Greene) K. Sch. (*Laciniaria Tracyi* Alexander). Occasional from North Carolina to Florida; abundant from Tennessee to Alabama, and again occasional westward to Indiana, Louisiana, and Texas.



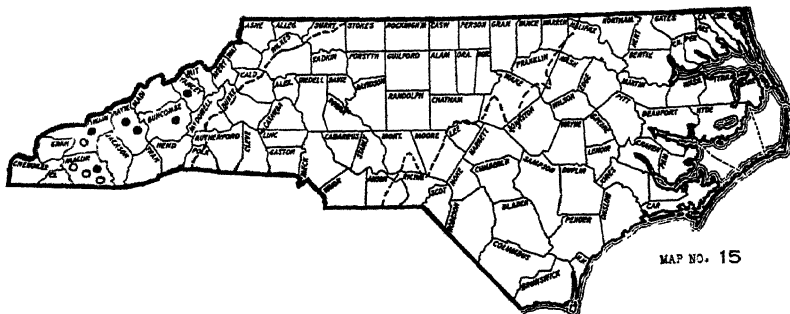
MAP NO. 13

MAP No 13. *Liatris aspera* Michx., var. *intermedia* (Lunnell) Gaiser. Occurring east of the Mississippi River through North and South Carolina to Georgia and Florida; from Ontario through Indiana and Kentucky to Alabama and west of the Mississippi more generally in the south central states.



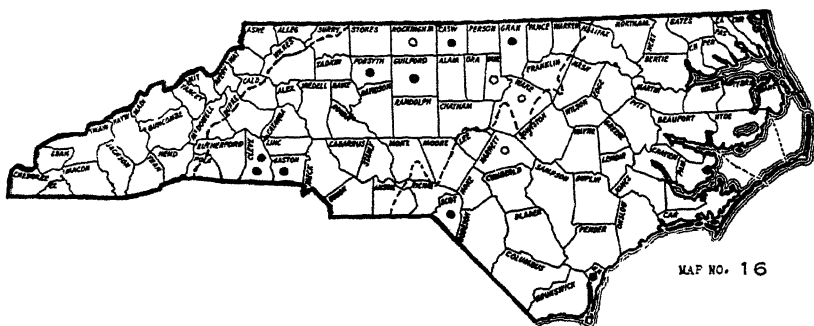
MAP NO. 14

MAP No 14. *Liatris scariosa* (L.) Willd., var. *squarrosula* (Michx.) Gray. Piedmont and coastal plain of North and South Carolina, Georgia, and Alabama.

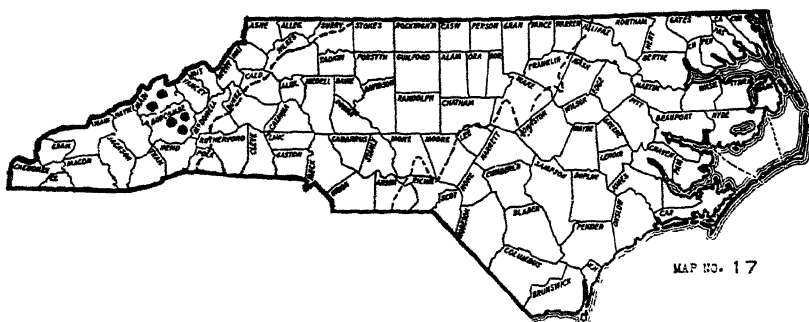


MAP NO. 15

MAP No 15. *Liatris scariosa* (L.) Willd., var. *virginiana* (Lunnell) Gaiser. Mostly in the mountains from southern Pennsylvania to South Carolina.



MAP No 16. *Liatris squarrosa* (L.) Michx., var. *typica* Gaiser. Delaware to northern Florida, westward to Alabama, Illinois, and Missouri.



MAP No 17. *Liatris squarrosa* (L.) Michx., var. *gracilentia* Gaiser. Mostly from southern Georgia to Florida, Alabama, Mississippi, and Louisiana.

NOTES ON FIELD OBSERVATIONS OF CERTAIN SPECIES AND VARIETIES

When collecting *Liatris* in the coastal plain I have noted that there are rather remarkable habit-habitat correlations which are not so evident when studying herbarium specimens. This may be true also for other regions and other species. I have been guilty too often, I find, of collecting selected small specimens without so indicating on labels, and perhaps other collectors do similarly.

One may, from seeing all the herbarium specimens of *L. secunda*, for example, get an entirely erroneous impression of the habit of the plant as it grows in the field in certain habitats. This species in southeastern North Carolina is a plant of the coarser sands, individual plants occurring rather sparsely scattered in the long-leaf pine, scrub oak barrens. This, I presume, is its natural habitat, and in this situation the plants are relatively slender with the inflorescence being a loose, secund raceme. Thus it is usually seen in the herbaria and thus described. This species, however, as are some of the others, is often found in great abundance in recently disturbed soils, particularly the plowed fire-control lanes, power-line transects, etc., where habitally it takes on quite a different aspect. The plants are, by and large, more vigorous and diffusely branched so that the inflorescence,

rather than being a loose, slender, secund raceme, is a diffuse panicle with the secund arrangement of the heads much less evident. This, apparently, is purely a vegetative growth response to a particularly favorable set of environmental conditions altering the more familiar aspect of the plants and no purely technical differences have been seen to correlate with it.

Liatis spicata, var. *resinosa*, as I have seen it, is naturally a plant of savannas, or of small grass-sedge bogs, where it is typically small, slender, and with a relatively short spike of heads. As such it is quite unlike variety *typica* of more northern regions and of the mountains in this state. It, too, invades the disturbed soils, where it flourishes to the extent that one would scarcely associate it with the savanna plant. It is generally much larger, with great long spikes, superficially resembling var. *typica*. In its more technical characters, however, like *L. secunda*, it seems to me to retain the nature of the variety *resinosa*. Dr. Gaiser cites a number of specimens from eastern North Carolina, among others, as intermediate between *L. spicata*, var. *typica* and var. *resinosa*. I venture to suggest that these cited intermediates are actually more vigorous and robust specimens of variety *resinosa*, and further that this variety may prove worthy of specific rank.

L. spicata, var. *resinosa* is described as "glabrous (hirsute only very exceptionally)." Dr. William B. Fox, collecting recently northeast of Lake Waccamaw in Columbus County, N. C., brought in some specimens of this variety all of which were pilose throughout, densely so at the nodes, otherwise not departing in character from the usual. I have since visited several localities in Brunswick and Columbus Counties where the variety was growing in abundance. In several places near Ash and Old Dock in Columbus County the populations of plants of both disturbed and undisturbed situations were about evenly mixed with plants that were quite glabrous and plants that were conspicuously pubescent as described above. In other respects they were similar. A number of localities visited near Southport in Brunswick County failed to have any individuals that were other than glabrous.

THE COASTAL PLAIN OF NORTH CAROLINA

BY H. W. STRALEY, III

Consulting Geologist and Geophysicist

Box 5532, Friendship Station, Washington, 16, D. C.

INTRODUCTION

About the middle of the last decade, interest developed in finding petroleum in the eastern seaboard states. Alabama, Florida, Georgia, and the Carolinas were examined by exploring parties. The drilling of several dry holes in adjacent states halted the program in North Carolina before it was well started. It was not until the end of the late hostilities could be foreseen that interest was rekindled.

As Richards (1947) so aptly stated, it was the success attained in Alabama, at Gilbertown in 1944, and Florida, at Sunniland the preceding year, that re-awakened interest. Three wells have been drilled in Maryland since the war, 12 in North Carolina, 21 in Georgia, and more than 40 in Florida. In the period embracing the early war years, before the entry of the United States, little effort was expended on coastal plain problems except by a handful of unsupported scientists.

GEOLOGY

The region has definite similarities with both the Gulf Coast and the Appalachian belt. Most of the tectonic features are subparallel with those of the Appalachian region, whereas the formations are those of the Mississippi Embayment but, in most instances, considerably attenuated. In the southern part of the Carolina plain there is one tectonic feature, however, that is discordant with the rest. The Carolina Ridge is transverse to Appalachian structure and subparallel with trends that are common in the post-Cretaceous rocks of Cuba.

The Cretaceous occupies a wide band inward from the coast, particularly along the Carolina Ridge. Coastwise and farther north, it is buried by later Tertiary and Quaternary formations. In the subsurface, both Tuscaloosa (basal Upper Cretaceous) and Potomac (Lower Cretaceous) thicken and become more marine toward the coast. It is sometimes difficult to distinguish between the two and they are often confused and mapped as one unit. Both Peedee and Black Creek (Navarro and Taylor, Upper Cretaceous) pinch out against the Carolina Ridge from their outcrops south of the Neuse River, as well as against the Fort Monroe elevation to the northeast.

The Eocene, which overlies them unconformably, pinches and swells in like manner. Wilcox is found in the subsurface; Claiborne is reported to outcrop near Clayton, Garner, Lillington, and Raleigh; and Castle Hayne (Jackson) is found in the outcrop along the Carolina Ridge and south of the Neuse River. It continues over the arch and becomes the Cooper-Santee formation in South Carolina.

An irregular basement complex, which slopes gently toward the sea, underlies the sedimentary cover. That peneplanation is far from complete is recognized at Fountain, Pitt County, where 60 meters of basement relief brings granite to the surface. Johnson (1938) described a valley and ridge, with about 100 meters of relief, between the fall zone and the longitude of Plymouth and Edenton. Sediments over a rolling topography may be compacted into supratenuous folds which may form local oil or gas traps.

Two distinct slopes for the surface of the basement complex have been recognized. Between the fall zone and Beaufort, Carteret, and Washington counties the gradient is about 0.2 meters per kilometer (Johnson, 1938; Miller, 1937). West of Fort Landing, Swanquarter, and Elizabeth City it steepens notably. Miller has attributed this to warping of the Paleozoic continent of Appalachia, but other interpretations appear to be equally probable and will be considered in a forthcoming paper by the present author.

A number of deep wells have been drilled in recent years. In 1945, the Tidewater Petroleum and Gas Company bottomed in basement complex at 1230 meters, after drilling 33 meters into the crystalline rock. A second well, in Pamlico Sound, was still in Cretaceous at 1929 when abandoned. No commercial oil or gas has been discovered in any of the wells drilled in the state.

MacCarthy and Straley (1936, 1937) investigated an area near the Carolina Ridge in 1935 and 1936. They mapped a number of magnetic anomalies, which they interpreted as basement structures, subparallel with the Appalachians and plunging to the southeast.

The Carolina Ridge, itself, may be interpreted in terms of topography or structure. The basement rocks are within 425 meters of the surface at Wilmington and both geology and geophysics indicates a northwestward extension of the crystalline mass. The Eocene and other formations, cited above, may feather out against a high ridge of resistant rock standing above the pre-Cretaceous erosion surface. On the other hand, the arch may represent folding of Cretaceous age equivalent to the northwest-southeast deformation in the West Indies.

One oil company considered drilling the Ridge in the 1930's, but abandoned the project. The light cover, the uncertainty of the section, and the probability that the sediments deposited there are of shallow water origin decrease its promise.

GEOPHYSICAL WORK

Several geophysical methods have been applied successfully to the Atlantic Coastal Plain. In general, the work has been carried out by four groups. North of the Dismal Swamp, Lehigh and Princeton universities have conducted gravitational, magnetic, and seismic surveys. In North and South Carolina, the University of North Carolina has played the leading role. The Department of the Interior made 16 magnetic profiles across the Florida peninsula not long before the outbreak of hostilities in Poland, in 1939. Oil companies have worked parts of the plain in considerable detail.

Most of these workers have agreed on certain dominant features. There is a sharp change of gradient near the coast, which is reflected on all profiles. Most of the structures indicated have a trend that links them with Appalachian tectonic features; the Carolina Ridge is the only exception north of South Carolina. The geophysical anomalies are due more to differences in lithology of the basement rocks than to topographic elevations or depressions in its surface.

In North Carolina, there are two notable exceptions to this unanimity. Johnson interpreted his findings in northeastern counties to indicate topographic relief of the order of 100 meters. An undefined magnetic anomaly along the Cape Fear River may be interpreted as representing the nearness of the crystalline rocks to the surface of the coastal plain rather than to basement lithology.

PETROLEUM

The writer is of the opinion that the Atlantic Coastal Plain should not be too heavily discounted until exploration has been completed. Among the areas that deserve more thorough investigation is the Pamlico basin lying between the Dismal Swamp and the Carolina Ridge.

REFERENCES

- JOHNSON, W. R., JR. 1938. Geomagnetic reconnaissance on the coastal plain of northeastern North Carolina. *GSA Bull.* **49**: 1951.
- MACCARTHY, G. R. 1936. Magnetic anomalies and geological structures of the Carolina coastal plain. *Jour. Geol.* **44**: 396-406.
- AND H. W. STRALEY, III. 1937. Magnetic anomalies near Wilmington, N. C. *Science* **85**: 362-364.
- AND ——— 1938. Geomagnetic reconnaissance of the Carolina coastal plain. *GSA Bull.* **49**: 1953.
- MILLER, B. J. 1937. Geophysical investigations in the emerged and submerged Atlantic coastal plain, Part II: Geological significance of geophysical data. *GSA Bull.* **48**: 803-812.
- RICHARDS, H. G. 1945. Subsurface stratigraphy of the Atlantic coastal plain between New Jersey and Georgia. *AAPG Bull.* **29**: 885-956.
- 1947. The Atlantic coastal plain: its geology and oil possibilities. *World Oil* **127**: 48.

SOUTHERN CHEMISTS: THEIR TRAINING AND CAREERS

By WILL S. DeLoach

*Huntingdon College
Montgomery, Alabama*

Where do Southern chemists get their training, where do they settle after their schooling is finished, and into what types of work do they go? This investigation was undertaken in order to answer these and certain related questions.

The chemists and chemical engineers listed in the seventh edition of "American Men of Science"¹ were chosen as a representative group high in their profession and relatively easy to study. Data were taken for all persons who listed "chemistry" or "chemical engineering" as their field, or one of their fields, and who were born in the following states: Alabama, Arkansas, Florida, Georgia, Louisiana, Mississippi, North Carolina, South Carolina, Tennessee, Texas, and Virginia, and received their undergraduate training there. State and date of birth, school and date of bachelor's and doctor's degrees, and last location and type of work listed were recorded. Because of the time necessary to compile and print a work like "American Men of Science", the figures on location and type of work actually represent the situation as it was some time prior to 1944. At that time, of course, many chemists were being affected by the war, as to where they were and what they were doing. The 1947 Directory of the American Chemical Society was consulted in order to find the 1947 addresses of those members of this group who are listed there.

For purposes of comparison, the same group in the first edition of "American Men of Science"², published in 1906, was studied, and the same information was collected. In the first edition 38, and in the seventh 603 were found who met the requirements previously stated. Since the former listed about 4,000 scientists and the latter about 34,000, it can be seen that the percentage of Southern chemists increased in about forty years from less than 1% to nearly 2% of the total number of scientists listed.

Table IA shows the native states for the two groups, and brings out certain trends. The most spectacular of these is the rise of Texas from close to last to first. In all, the fact that seven of the states contribute a greater percentage in the second group indicates the development of an interest in chemistry, and, probably, the increase in educational opportunities in those sections. Virginia and North Carolina are in the top three in both groups, but contribute a smaller fraction of the total in the second. In 1906, almost half were natives of these two states.

¹ Cattell, Jaques, "American Men of Science", 7th ed., The Science Press, Lancaster, Pa. 1944.

² Cattell, Jaques, "American Men of Science", 1st ed., The Science Press, New York, 1906.

Table IB shows the number of native chemists per 100,000 population in the various states. Census figures for 1900 and 1940 were used. The eastern seaboard states dominate both groups, with Virginia, North Carolina, South

TABLE IA

Native states

1906 GROUP			1944 GROUP		
State	Number of native chemists	% of total	State	Number of native chemists	% of total
Virginia.....	11	28.9	Texas.....	111	18.4
North Carolina.....	7	18.4	Virginia.....	107	17.7
South Carolina.....	5	13.2	North Carolina.....	84	13.9
Mississippi.....	4	10.5	Georgia.....	59	9.8
Georgia.....	3	7.9	Tennessee.....	52	8.7
Alabama.....	2	5.3	South Carolina.....	42	7.0
Tennessee.....	2	5.3	Alabama.....	39	6.5
Texas.....	2	5.3	Mississippi.....	36	6.0
Arkansas.....	1	2.6	Arkansas.....	30	5.0
Louisiana.....	1	2.6	Louisiana.....	28	4.6
Florida.....	0	0	Florida.....	15	2.5
Totals.....	38		Totals.....	603	

TABLE IB

Native chemists per 100,000 population

1906 GROUP				1944 GROUP			
State	Pop.	Native chemists	Chem. per 100,000 pop.	State	Pop.	Native chemists	Chem. per 100,000 pop.
	<i>hundred thousands</i>				<i>hundred thousands</i>		
Virginia.....	19	11	0.58	Virginia.....	27	107	3.92
S. Carolina.....	13	5	0.39	N. Carolina.....	36	84	2.33
N. Carolina.....	19	7	0.37	S. Carolina.....	19	42	2.21
Mississippi.....	16	4	0.25	Georgia.....	31	59	1.90
Georgia.....	22	3	0.14	Tennessee.....	29	52	1.79
Alabama.....	18	2	0.11	Texas.....	64	111	1.73
Tennessee.....	20	2	0.10	Mississippi.....	22	36	1.64
Arkansas.....	13	1	0.08	Arkansas.....	19	30	1.56
Louisiana.....	14	1	0.07	Alabama.....	28	39	1.39
Texas.....	30	2	0.07	Louisiana.....	24	28	1.17
Florida.....	5	0	0	Florida.....	19	15	0.79

Carolina, and Georgia occupying the first four places in 1944. However, the proportionate increase of most of the other states was greater, which again points to the development of the other sections. In 1944, the state with the smallest number of native chemists per 100,000 population had more than the state that had the most in 1906.

The 38 members of the 1906 group attended 23 different schools for undergraduate degrees, while the 603 members of the 1944 group attended 110. Table II shows the location of these schools by states, and the number of graduates from the schools in each state. Again the early pre-eminence of the eastern seaboard states and the later development of other sections is illustrated: 73% of the first group received bachelor's degrees from schools located in Virginia, North Carolina, South Carolina, or Georgia, as compared to 51% of the later group. The spreading out of educational opportunity may also be illustrated by the fact that 73% of the first group received bachelor's degrees from schools located in their native states, as compared to 82% for the later group.

The chances are that a member of the 1944 group received his bachelor's degree from a school, most probably a university, that was coeducational, that

TABLE II
Location of undergraduate schools

1906 GROUP				1944 GROUP			
State	No. of grad.	% of total	No. of schools	State	No. of grad.	% of total	No. of schools
Virginia . . .	13	34.2	7	Texas	112	18.6	20
N. Carolina . . .	8	21.1	4	Virginia	112	18.6	15
Georgia . . .	4	10.5	1	N. Carolina . . .	96	15.9	14
S. Carolina . .	3	7.9	3	Georgia	55	9.1	9
Tennessee . . .	3	7.9	3	Tennessee . . .	51	8.5	19
Alabama	2	5.3	1	S. Carolina . . .	44	7.3	8
Louisiana . . .	2	5.3	2	Alabama	34	5.6	6
Texas	2	5.3	1	Mississippi . . .	32	5.3	7
Mississippi . . .	1	2.6	1	Arkansas	27	4.5	4
Arkansas . . .	0	0	0	Louisiana	24	4.0	5
Florida	0	0	0	Florida	16	2.7	2
Total	38		23	Total	603		110

operated under public control, and that had a relatively large student body, judging by the 1940-1941 enrollment. It is worth noting, however, that many liberal arts colleges with small enrollments contributed heavily. Eight colleges whose 1940-1941 enrollments ranged from 309 to 505 have a total of 50 graduates in the group. Probably the outstanding example of this is found in Mississippi—of the 32 who received their bachelor's degrees in that state, 13 come from a small denominational college whose 1940-1941 enrollment was 505. If the individual attended a church-controlled school, as about 39% did, the odds are four to one that it was Baptist, Methodist, or Presbyterian. It may be a little surprising to learn that technical schools contributed less than 10% of the total. Two schools, Alabama Polytechnic Institute, with 21, and North Carolina State, with 12, account for nearly two-thirds of the contribution of technical schools. Students are getting older at graduation. In the first group, the age ranges from 17 to 30, with 72% being graduated at 21 or younger, while in the second group the range is from 17 to 43, with 53% being graduated at 21 or younger.

Of the 110 schools which members of the 1944 group attended for undergraduate work, 19 are on the December 25, 1946, list of schools that have chemistry departments approved by the American Chemical Society for professional training.

These 19 schools account for 294, about 49%, of the total. With one exception, every Southern state has at least one chemistry department approved by the A. C. S. represented in this list, and Virginia has four, including the only one in this group in a woman's college.

Table III summarizes the data regarding the undergraduate work for the 1944 group, and, for purposes of comparison, gives some data on the earlier group.

TABLE III
Nature of undergraduate schools

TYPE OF SCHOOL	1906 GROUP			1944 GROUP		
	No. of schools	No. of graduates	% of graduates	No. of schools	No. of graduates	% of graduates
			%			%
College	12	18	47.4	58	214	36.5
University.	8	15	39.5	26	306	52.1
Technical.	3	5	13.1	8	54	9.2
Teacher's col.	0	0		5	13	2.2
Control						
Public.	12	22	55.6	34	311	52.5
Private, non-sect.	3	3	8.3	13	52	8.8
Denominational.	8	13	36.1	54	229	38.7

1944 GROUP

Nature of student body	No. of schools	No. of graduates	% of total	Size of student body (1940)	No. of schools	No. of graduates	% of total
Co-ed	78	441	75.1	Fewer than 500	27	74	12.8
Men	16	136	23.2	500 to 1500	40	178	30.6
Women	6	10	1.7	More than 1500	29	329	56.6

In the 1906 group 68%, and in the 1944 group 84% listed doctor's degrees that were presumably earned, since information to the contrary was not given. Table IV presents a list of schools granting such degrees to the members of the 1906 group and a partial list for the other group. In the latter case, all the Southern and all the foreign schools are listed, as well as all the other schools represented by five or more doctorate alumni. Prior to 1906, Johns Hopkins was the overwhelming favorite, with 58% of the doctor's degrees being granted there. Only two out-and-out Northern schools are represented, and those by only one alumnus each. The picture is very different in 1944. Johns Hopkins still leads, with 58; but Virginia, with 45, and North Carolina, with 37, are close behind, and Texas ties Chicago at 29. Over 50 schools have granted 506 doc-

tor's degrees to members of this group, the earliest in 1880, the latest in 1943. That span of approximately sixty years falls naturally into three periods. During the first twenty years, only an occasional Ph.D. is found—actually, six

TABLE IV
Schools granting doctor's degrees

1906 GROUP		1944 GROUP	
University	No. of graduates	University	No. of graduates
Johns Hopkins	15	Johns Hopkins	58
Gottingen	2	Virginia	45
North Carolina	2	North Carolina	37
Virginia	2	Chicago	29
Cornell	1	Texas	29
Missouri	1	Columbia	25
Munich	1	Yale	20
Pennsylvania	1	Cornell	19
Vanderbilt	1	M. I. T.	18
		Duke	15
		Iowa State	15
		Ohio State	15
		Iowa	14
		Illinois	13
		Wisconsin	13
		Princeton	10
		Calif. Inst. Tech.	8
		Harvard	8
		Purdue	8
		California	7
		Penn. State College	6
		Cincinnati	5
		Colorado	5
		Florida	5
		Northwestern	5
		Pittsburgh	5
		Rice	5
		Vanderbilt	4
		L. S. U.	3
		Peabody	3
		Gottingen	2
		Berlin	1
		Besancon	1
		Leipzig	1
		N. C. State	1
		T. C. U.	1
		Washington and Lee	1

prior to 1900. During the next twenty years there is at least one every year, and there are several groups of five. World War I saw a new high: from 1914 through 1917 the average was six a year. The results of our entrance into the war show in these figures: two for 1918, and one for 1919. From 1920 on there

are at least six a year, except for 1943, when there were four. A new high level was reached during 1929-1932; the average for that period was 27.5 a year. The effects of the depression were not noticeable until 1933, when the number

TABLE V

Location

1906 GROUP		1944 GROUP	
Location in 1906	No.	Location in 1944	No.
North Carolina	6	New York.....	60
Georgia	3	Texas.....	48
Louisiana.....	3	Virginia.....	44
Mississippi	3	New Jersey.....	39
Tennessee.....	3	North Carolina	36
Texas.....	3	Pennsylvania.....	32
Alabama.....	2	Georgia	29
South Carolina.....	2	Alabama.....	24
Virginia.....	2	Washington, D. C.....	24
Washington, D. C.....	2	Louisiana.....	23
Connecticut.....	1	Delaware	22
Indiana.....	1	Illinois	21
Kentucky.....	1	Maryland.....	20
Maryland.....	1	South Carolina.....	20
Missouri.....	1	Tennessee	17
New York.....	1	Ohio.....	16
Ohio.....	1	California	15
Pennsylvania.....	1	Indiana.....	13
San Juan, P. R.....	1	Mississippi.....	12
		Connecticut	10
		Florida	10
		Michigan	9
		Massachusetts.....	8
		Missouri.....	8
		West Virginia	8
		Arkansas	7
		Wisconsin	7
		Oklahoma.....	6
		Minnesota.....	5
		Arizona	2
		Iowa.....	2
		Kansas.....	2
		Colorado.....	1
		Kentucky.....	1
		Oregon	1
		Wyoming	1

fell to 16. The period 1938 through 1941 set another high mark, and has an average of 30 a year.

Although the age of those receiving this degree ranges in the 1944 group from 21 to 54, the most popular time is the middle twenties, about 40% being 25, 26,

or 27 years old. While the number decreases with age rather steadily from the peak at 26, there is a sharp drop at age 38, and the number receiving the degree at that age or older amounts to about 4% of the total. The figures for the 1906 group agree fairly well; the range is from 23 to 39, and the highest level is reached at 25 and 26.

TABLE VI
Kind of occupation

OCCUPATION AT TIME OF REPORT	1906 GROUP		1944 GROUP	
	Number	%	Number	%
Academic	31	81.6	261	43.3
Government	5	13.2	63	10.4
Industry	2	5.3	252	41.8
Military service			20	3.3
Other			7	1.2
Total	38		603	

TABLE VII
Occupations of those located in the South

STATE	1906 GROUP				1944 GROUP					
	Aca- demic	Gov't.	In- dustry	Total	Aca- demic	Gov't	In- dustry	Mil. Serv.	Other	Total
Alabama	2			2	15	6	2	1		24
Arkansas				0	6	0	1	0		7
Florida				0	6	1	2	0	1	10
Georgia	3			3	23	1	4	1		29
Louisiana	3			3	9	5	8	1		23
Mississippi	2	1		3	10	0	2	0		12
N. Carolina	5	1		6	23	3	9	1		36
S. Carolina	2			2	15	0	2	3		20
Tennessee	3			3	13	1	3	0		17
Texas	3			3	32	1	14	1		48
Virginia	2			2	25	6	11	2		44
Totals	25	2	0	27	177	24	58	10	1	270

Six members, about 1%, of the 1944 group were singled out, by being "starred", as outstanding in their fields. Of this number, only one—a South Carolinian doing industrial work in his native state—was located in the South. None of these are young; their ages in 1944 ranged from 65 to 89. Of the 1906 group, 4—about 10%—were "starred", and two were in the South at that time, both in academic work.

In 1906, the chances were that a member of that group was engaged in teaching, as were about 82%, and that he was located in the South, as were about 71%. Of the 27 in the South, only two were not in academic work, and they were employed by state governments.

In 1944, about 43% were in academic work, about 10% were employed by a state or by the national government, nearly 42% were in industrial work, and a little over 3% were in military service. This last number is probably too small, but only those about whom there was no doubt were recorded as being in military service. Three hundred and thirty-three, or about 55%, were located out of the South. Of those in the South, nearly 66% were in academic work, nearly 9% were in government service, about 21% were in industry, and nearly 2% were in military service.

The 1947 Directory of the American Chemical Society lists 495 members of the 1944 group. Of this number, 288, or about 58%, were located out of the South.

Table V gives the locations of the members of the two groups, Table VI their occupations, and Table VII gives the occupations of those located in the South.

Some observers have expressed concern with the effects on the South of the continual loss of its human resources to other regions. Douglas Southall Freeman, eminent Southern editor and author, giving a commencement address at a Southern college in August, 1947, stated that while he did not consider the South to be the nation's No. 1 economic problem, he did feel that "if we could bring back just one-third of the ablest men and women which the Southern region has lost to the North, they would form a cadre for the building of a better South." Others have pointed out that the movement of large numbers of Southerners to other parts of the country represents a grave economic loss, because of the cost of rearing and educating those that leave.

There is no doubt that the different regions and the country as a whole profit from the free intermingling of the population. The South is better off from having some of its chemists go to other regions and chemists from other parts of the country settle in the South. However, a net loss for a given region over the years is a different matter. A region is bound to suffer if there is a continual drainoff of its more competent and better educated citizens. It is not the purpose of this study to determine whether, over a period of time, the chemists who come to the South from other regions replace those that leave, but that is a point that might be investigated. The facts are that in 1906 about 29%, in 1944 about 55%, and in 1947 about 58%, of the South's chemists had gone elsewhere. Furthermore, in 1944 five of the six considered to be outstanding were located out of the South.

As long as more, and possibly better, opportunities exist elsewhere, Southern chemists will continue to seek them. However, when the South is able to offer enough attractive positions in chemistry and chemical engineering, this exodus should decrease. The continued development of good graduate departments in some Southern universities, the establishment of government projects such as that at Oak Ridge and of research institutes like those in Birmingham and Chattanooga, and the growth of chemical industries such as those in southwestern Louisiana and eastern Texas indicate that in the future the South will be a more attractive and profitable location for the chemist and chemical engineer.

A NEW SPECIES OF ACHLYA AND OF DICTYUCHUS¹

By E. S. BENEKE

*Department of Botany, University of Illinois
Urbana, Illinois*

PLATES 29 AND 30

During the progress of a study of numerous collections of water molds occurring in the state of Illinois, two belonging to the Saprolegniaceae were found which are apparently not identical with any which have been described previously. The first was isolated on *Crataegus* twigs from the Desplaines River and belongs to the genus *Achlya*. The other, isolated from soil and water samples from several streams in the state, is a new species of *Dictyuchus*.

Both of these water molds were grown as single hyphal and single spore cultures so that their characteristics could be carefully studied in unifungal culture. The following descriptions were drawn up from cultures growing in Petri dishes on sterilized hemp seed in charcoal-treated, sterilized, distilled water.

***Achlya intricata* n. sp.**

PLATE 29

Mycelial growth moderately dense, reaching a length of about 1.4 cm. on hempseed in a Petri dish. Hyphae mostly 18–35 μ in diameter, up to 133 μ at the base, branching considerably, tapering to a bluntly pointed tip. Gemmae abundant, formed in rows by segmentation, usually more or less cylindrical or ovoid, a single gemma sometimes measuring up to 1250 μ in length, frequently with knobs or projections at the ends, upon germination forming thin hyphae that usually develop small sporangia on their ends. Sporangia moderately abundant, subcylindrical to long slender, frequently tapering toward the tip, about the same width as the hyphae, variable in size, measuring 18–36 μ by 186–445 μ , these emptying either as is usual for *Achlya*, or remaining within sporangia, germinating *in situ*; spores 10–12.5 μ in diameter, forming a rather loose sphere about the mouth of the sporangium. Oogonia fairly abundant, almost always borne terminally on the main hyphae, but sometimes on very long lateral branches. Oogonia very irregular in shape, long-cylindrical to spherical, sometimes lacking lateral outgrowths or papillate projections, but usually having prominent outgrowths that as a rule contain one or more eggs, the larger outgrowths sometimes reaching the size of smaller oogonia, and becoming separated by a cross wall to give the appearance of an irregular chain of oogonia. Oogonial measurements extremely variable, cylindrical oogonia about 18–35 μ by 75–208 μ , ovate to spherical oogonia about 35–45 μ by 55–65 μ excluding the outgrowths, which may extend up to 95 μ in length. Oogonial wall smooth, unpitted except under antheridia; oospores normally spherical, or shape may be altered by position, eccentric, 4–20 or more, 17–31 μ , averaging about 20–23 μ in diameter, elongated ones up to 48 μ in length. Antheridial branches numerous,

¹ The writer wishes to express his sincere appreciation to Dr. Leland Shanor for his helpful suggestions during the investigations and the preparation of this paper.

diclinous, long and much-branched, in many cases coiling about the oogonial hyphae and oogonia, usually more than one to each oogonium; antheridial tubes not seen.

Collected once growing on *Crataegus* twigs in Desplaines River near Dam No. 1, four miles south of Wheeling, Ill., October 7, 1947. Slides from type culture deposited in Mycological Collections, University of Illinois Herbarium.

Myceliis in semine *Cannabis sativae* densis, quandoque usque ad 1.4 cm. longis; hyphis in basi ad 133μ , plerumque inter 18 et 35μ diam., ramosis. Gemmis singularibus aut ordine dispositis, cylindratis, ex articulis hypharum efformatis, que germinantes multa sporangia emittunt. Sporangii sub-cylindratis, huic generi propriis, inter 186 et 445μ longis; sporis inter 10 et 12.5μ diam. Oogoniis terminalibus, inaequaliter formatis, cylindratis sive globosis; papillis sive excrescentibus, usque ad 95μ longis; oogoniis cylindratis, inter 18 et 35μ diam. et inter 75 et 208μ longis, oogoniis globosis, inter 35 et 65μ diam. Tunica oogoniorum nullis punctulis notata et omnino levi; 4–20 oosporis, excentricis, inter 17 et 31μ diam., sed plerumque inter 20 et 23μ diam.; in papillis natis. Antheridiis origine dicliniis. Hab. in ramulis deiectis *Crataegus* spp., Desplaines River, Wheeling, Illinois.

This species is quite distinct from other known species of *Achlya* because of the development of very irregular oogonia with one or more eggs usually present in the prominent outgrowths. Two species, which may have outgrowths, were reported by Harvey in 1942, but these two species only occasionally have projections on the oogonia. The cylindrical, intercalary oogonia of *Achlya pacifica* Harvey may have numerous germ-tube-like projections. These extensions are never very long and oospores do not develop in them, contrasting distinctly to the condition common in the present species. *Achlya pinnulata* Harvey usually has spherical oogonia, although on rare occasions irregularly lobed oogonia with one or two oospores in each lobe may be found. The presence of oospores in the lobes of the oogonia in *A. pinnulata* is the nearest approach to the condition found regularly in *A. intricata*. However, the oogonia of *A. pinnulata* are usually spherical, the lobes which are rarely developed are not as long or prominent.

Achlya recurva Cornu (Cornu, 1872; Latham, 1935) has spherical oogonia with numerous protuberances up to 16μ long, and sometimes has a few atypical oogonia that are irregular in shape, with enormously elongated protuberances. These protuberances are quite different from those of *A. intricata* in that the length is not as great, the longer ones are atypical, and the ends are truncate and thin-walled in *A. recurva*.

The eccentric nature of the oospores and the diclinous origin of the antheridia would place *A. intricata* in the "Prolifera Group" (Coker, 1923). Although species of this group usually lack or have only a few papillae on the oogonia, the other characteristics of this group certainly would indicate that *A. intricata* belongs here. *Achlya intricata* differs from other members in the "Prolifera Group" by the development of extensive outgrowths which result in very ir-

regularly-shaped oogonia that vary from more or less spherical to cylindrical in form. These protuberances usually contain one to many eggs.

Dictyuchus pseudoachlyoides n. sp.²

PLATE 30

Mycelium fairly vigorous on hempseed, reaching a length of 1.3 cm.; hyphae up to 60μ thick at base, mostly about $35\text{--}45\mu$ in diameter. Gemmae formed abundantly in week-old cultures, usually intercalary, often immediately below sporangia, solitary or in chains, spherical, oval, or club-shaped. Primary sporangia borne on tips of the main hyphae, secondary ones formed by cymose branching; a few of the early sporangia, under favorable conditions, discharge spores as in *Achlya* although the majority of the sporangia are of the false-net type, in older cultures only the false-net type sporangia have been found; sporangia cylindrical, usually a little larger in the distal half, $14\text{--}32\mu$ by $93\text{--}562\mu$, mostly $25\text{--}30\mu$ by $250\text{--}350\mu$; spores $10\text{--}13\mu$ in diameter, of the same size in both types of sporangia, and in both cases emerging from their cysts and swimming once, or occasionally sprouting *in situ*. Oogonia abundant, spherical to sub-spherical, $28\text{--}41\mu$ in diameter, mostly 30μ , the walls unpitted, borne on long thin lateral stalks arising from the main hyphae, varying in length from about two to three times up to six or eight times the diameter of the oogonia, stalks straight or somewhat curved; eggs spherical, one to an oogonium, eccentric, $23\text{--}35\mu$ in diameter, averaging about 26μ ; antheridia on nearly all oogonia, androgynous, one to four arising from the oogonial stalk, very rarely from the main hyphae, antheridial branches which do not form antheridia frequently developing sporangia, or in some cases, secondary oogonia with accompanying antheridia.

Collected three times, once from sandy soil on bank of the Little Wabash River, Louisville, Ill., April 7, 1947 (type), once from water in the Beaver Creek, east of Springeton, Ill., April 9, 1947, and once from soil on bank of Deer Creek 5 miles east of Lincoln, Ill., May 3, 1947. Slides of type culture have been deposited in the Mycological Collections, University of Illinois Herbarium.

Myceliis quandoque usque ad 1.3 cm. longis; hyphis basi ad 60μ , plerumque inter $35\text{--}45\mu$ diam. Gemmis plus minusve copiose evolutis, globosis aut ovatis, frequentissime sub sporangiis, vel intercalariis. Sporangii terminalibus, secundariis sporangiis modo cymosis crescentibus. Aliquibus sporangiis *Achlya* instar, plerumque falsireticulis formis, $14\text{--}32\mu$ lat. et $93\text{--}452\mu$ long., plerumque $25\text{--}30\mu$ lat. et $250\text{--}350\mu$ long. Sporis $10\text{--}13\mu$ diam. Oogoniis globosis, $28\text{--}41\mu$, plerumque 30μ diam., tunica oogoniorum non-punctulata, in ramulis longis. Oosporis 1, guttulis oleosis excentrice coadunatis, $23\text{--}35\mu$, plerumque 26μ diam. Ramulis antheridiorum 1-4, androgenis, e ramulis oogonialibus, sub oogoniis nascentibus. Aliquibus ramulis antheridiorum cum sporangiis aut secundariis oogoniis. Hab. ad terram umidam, Little Wabash River, Louisville, Illinois.

This species may be distinguished readily from the others in the genus *Dictyuchus* because it possesses smooth spherical oogonia which are accompanied by

² The writer wishes to acknowledge with appreciation the assistance of Dr. F. D. Lazenby, Department of Classics, University of Illinois, in the preparation of the Latin diagnoses.

numerous androgynous antheridia which arise from the oogonial stalk. The phenomenon of sporangial or oogonial formation on antheridial branches is interesting and not infrequently encountered.

Dictyuchus pseudoachlyoides has characteristics that relate it more closely to *Dictyuchus achlyoides* Coker & Alexander (Coker, 1927) than to any other species of *Dictyuchus*, but it differs from *D. achlyoides* in several important features. *Dictyuchus achlyoides* possesses spherical oogonia with many papillae 3–10 μ long, and the antheridia, when present, usually occur singly, being on approximately 65 per cent of the oogonia. Oogonia of *D. pseudoachlyoides*, in contrast, lack papillae, two to four antheridial branches are usually present, which arise from the oogonial stalk, and antheridia are associated with nearly all of the oogonia. The formation of gemmae in young cultures and the location of many of these immediately below the sporangia is also a conspicuous character.

Dictyuchus monosporus Leitgeb (Couch, 1926) is readily separated from *D. pseudoachlyoides* in that the former is typically heterothallic, although frequently represented in collections by only one of the neutral or compatible strains, develops a true net in the sporangia, and lacks the *Achlya* type sporangia, while the latter is always homothallic, antheridia arise androgynously from the oogonial stalk, and the *Achlya* type sporangia may develop.

Dictyuchus missouriensis Couch (1931) lacks antheridia, frequently the oogonium is joined to the stalk by a beak-like process, and the distal half of the hyphae may have a zigzag-like arrangement because of the way the sporangia develop. These characteristics readily distinguish *D. missouriensis* from *D. pseudoachlyoides*.

Dictyuchus pseudodictyon Coker & Braxton (Couch, 1931) is distinctive from *D. pseudoachlyoides*, as the former has short oogonial stalks, androgynous or diclinous antheridia which may frequently almost enwrap the oogonium, apparently lacks gemmae, and the sporangia are long, thin, and very often complicatedly branched. The presence of gemmae, the *Achlya* type sporangia in young cultures, the lack of any diclinous antheridia, and the development of nearly all of the antheridia from the oogonial stalk in *D. pseudoachlyoides* are important differences.

SUMMARY

Two new species belonging to the Saprolegniaceae are described. The first species, *Achlya intricata*, is characterized by having very irregular oogonia with one or more eggs usually present in the extended outgrowths, sporangial formation typical of the genus, oogonia usually terminal, antheridia diclinous and frequently coiling antheridial branches around the oogonial hyphae and oogonia.

The second new species, *Dictyuchus pseudoachlyoides*, has spore behavior and sporangial formation typical of *Dictyuchus achlyoides*. This species is distinctive in having oogonia on long stalks, antheridial branches developing androgynously from the oogonial stalk, some antheridial branches bearing sporangia or secondary oogonia, and gemmae developing intercalarily or immediately below the sporangium.

LITERATURE CITED

- COKER, W. C. 1923. The Saprolegniaceae, with notes on other water molds. 201 pp. University of North Carolina Press. Chapel Hill, North Carolina.
- 1927. Other water molds from the soil. Journ. Elisha Mitch. Sci. Soc. 42: 207-226, pl. 36.
- CORNU, MAXIME. 1872. Monographie des Saprolegniées: étude physiologique et systématique. Ann. Sci. Nat. V, 15: 5-198.
- COUCH, J. N. 1926. Heterothallism in *Dictyuchus*, a genus of water moulds. Ann. Bot. 40: 849-881.
- 1931. Observations on some species of water molds connecting *Achlya* and *Dictyuchus*. Journ. Elisha Mitch. Sci. Soc. 46: 225-230, pl. 15.
- HARVEY, J. V. 1942. A study of western water molds. Journ. Elisha Mitch. Sci. Soc. 58: 16-42, pls. 4 and 5.
- LATHAM, D. H. 1935. *Achlya recurva* Cornu from North Carolina. Journ. Elisha Mitch. Sci. Soc. 51: 183-187, pl. 52.

PLATE 29

Figs. 1-12. *Achlya intricata*

Explanation of figures

Fig. 1. A typical sporangium. Fig. 2. A sprouted gemma with thin hyphae that develop small sporangia on their tips. Figs. 3, 4. Cylindrical type oogonia with declinuous antheridial branches entwining them. Figs. 5, 6. Gemmae. Fig. 7. An oogonium with eggs in the large prominent outgrowths. One of the outgrowths is cut off by a cross wall. Figs. 8-12. Typical irregularly shaped oogonia with one or more eggs in most of the prominent outgrowths. All drawings made with aid of a Spencer camera lucida. Fig. 1, $\times 258$; fig. 2, $\times 60$; figs. 3-5, $\times 258$; fig. 6, $\times 60$; figs. 7-12, $\times 258$.

PLATE 30

Figs. 1-13. *Dictyuchus pseudoachlyoides*

Explanation of figures

Fig. 1. False-net type sporangium showing spores emerging from their cysts. Fig. 2. Sporangium with spores sprouting in position. Fig. 3. Cymose branching of sporangia. Fig. 4. *Achlya* type sporangium. Figs. 5, 6. Intercalary gemmae in chains. Figs. 7, 8. Gemma immediately below sporangium and intercalary gemma. Fig. 9. Oogonium with antheridial branches, one developing into a sporangium. Fig. 10. Oogonium with four antheridial branches arising from oogonial stalk. Fig. 11. Oogonium with antheridial branches, one forming a secondary oogonium with accompanying antheridia. Fig. 12. Oogonium and antheridial branches, some terminated with spores. Fig. 13. Oogonium with antheridia. Figs. 1-2, $\times 258$; fig. 3, $\times 60$; figs. 4-6, $\times 258$; figs. 7-8, $\times 60$; figs. 9-13, $\times 258$.

PLATE 29

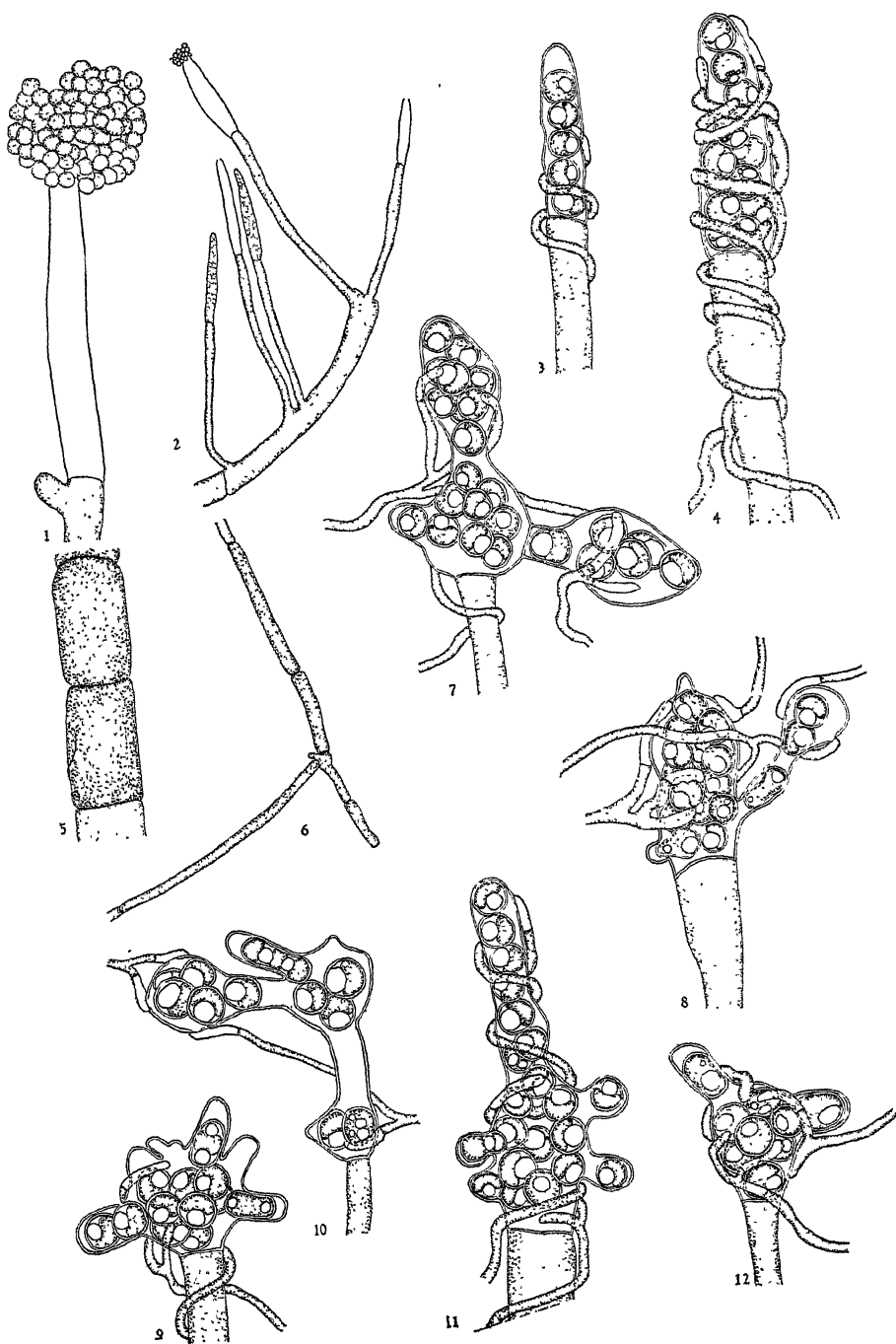
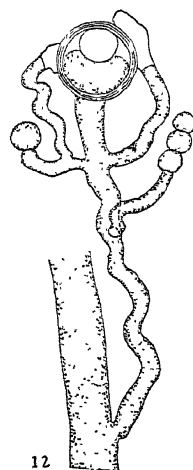
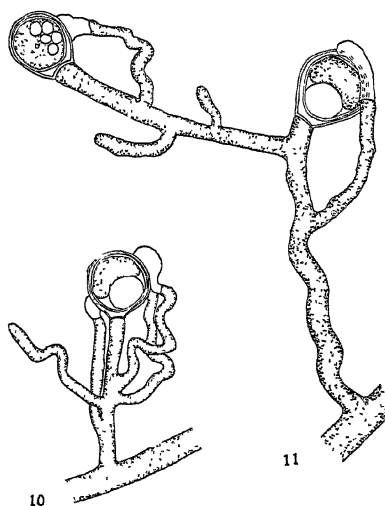
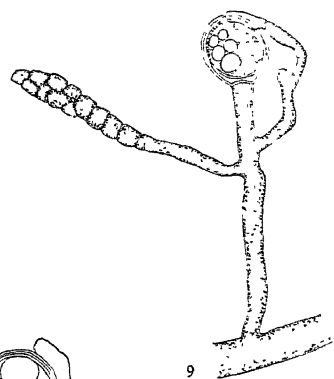
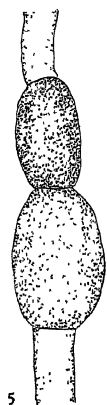
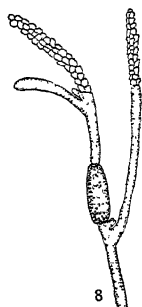
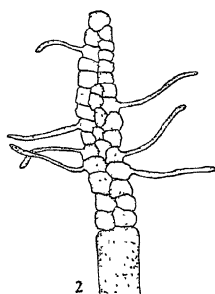
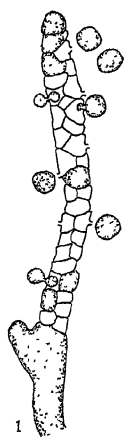


PLATE 30



STUDIES ON SOME WILD YEASTS FROM CHAPEL HILL AND VICINITY

BY HELEN S. SHERWIN

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina¹*

PLATES 31-34

In recent years wild yeasts have received increasing attention in this country. There has still been little work on them here, however, as compared with the extensive work done in Europe. Some of the recent investigations on yeasts in the United States are those carried on by Mrak and McClung (1940), Mrak, Phaff, and Smith (1942), and Mrak (1945) at the University of California. Bedford (1942), also working at the University of California, made a taxonomic study of the genus *Hansenula*. Nickerson (1943) described *Zygosaccharomyces acidifaciens*, a new acetifying yeast, and the same author (1944) reported on a study of the film-forming yeasts *Zygopichia* and *Zygohansenula*. Kurth (1946) reported on yeasts that were grown on still-waste liquor from the production of Douglas fir wood sugar alcohol at Springfield, Oregon, and Kurth and Cheldelin (1946) reported further work on this subject. Anderson and Skinner (1947) studied yeasts in decomposing fleshy fungi.

Some reports of yeasts from North Carolina have been made. Coker and Wilson (1911) first reported *Schizosaccharomyces octosporus* Beyerinck on the American continent. Conant (1940), of Duke University, published "The Taxonomy of the Anascosporous Yeast-like Fungi". Lehman (1943) reported on the occurrence of yeast spot on soybean in North Carolina.

Since there have been so few studies of the yeasts in this state, the present investigation was undertaken with the purpose of collecting, isolating, and identifying as far as possible the yeasts occurring on wild fruits, the bark, wood, and sap of trees, and in the soil from the vicinity of Chapel Hill. Two yeasts isolated from fungi and one yeast pathogen, a culture of which was turned over to the author, are also included in the study.

METHODS

Collections were made in the following manner: Samples of fruits, soil from underneath the plants, and fungi growing on dead wood were collected in separate envelopes. The samples were placed in sterile Petri dishes containing either potato-dextrose broth, beer wort, or distilled water. The fruits were crushed in order to enrich the liquid medium. Bark, wood, and sap were collected from trees in some of which a small cut had been made to allow the sap to flow out. These samples were put directly into sterile bottles containing one of the above

¹ Author's present address is Division of Forage Crops and Diseases, U. S. D. A. Plant Industry Station, Beltsville, Md.

liquid media, and were left in the bottles when they were brought into the laboratory.

Some of the collections made were the following: Fruits of hawthorn (*Crataegus*), privet (*Ligustrum*), firethorn (*Pyracantha*), dogwood (*Cornus*), honeysuckle (*Lonicera*), sumach (*Rhus*), wild rose (*Rosa*), barberry (*Berberis*), holly (*Ilex*), honey locust (*Gleditsia triacanthos* L.), *Elaeagnus*, *Symphoricarpos*, *Euonymus*, chinaberry (*Melia*), apple (*Malus*), horse nettle (*Solanum*), *Nandina*, persimmon (*Diospyros*), laurel cherry (*Laurocerasus*); dried grapes from underneath grape arbor; soil from underneath peach (*Prunus*), persimmon, (*Diospyros*), maple (*Acer*), and oak (*Quercus*) trees; a jelly fungus, *Exidia gelatinosa*, growing on a dead twig; *Sebacina* sp. growing on dead wood; decaying wood from a beech tree (*Fagus*); bark, wood, and some sap from red maple (*Acer rubrum* L.), sweet gum (*Liquidambar styraciflua* L.), beech, red cedar (*Juniperus*), sycamore (*Platanus*), Southern red oak (*Quercus palustris* Muench.), sugar maple (*Acer saccharum* Marsh); sap and growth on newly cut ironwood stumps (*Carpinus caroliniana* Walt.); and sap and growth on newly cut birch stumps (*Betula*). Not all of the collections yielded growths of yeasts.

The samples were left at room temperature in the Petri dishes and bottles. When noticeable growth occurred, it was examined microscopically. In most cases it was found to consist of bacteria and fungi of various kinds, sometimes including yeasts and budding cells which resembled yeast cells. *Penicillium* was collected frequently, particularly on the fruits.

The next procedure was the isolation of the yeasts in pure culture. Since the yeasts, as a rule, unlike the bacteria, prefer a slightly acid medium, growth of the two in such a medium will yield a more abundant growth of yeasts. Thus the first step in purification was the transfer of a loopful of the mixed growth in the liquid medium to a test tube containing 10 cc. of potato-dextrose broth and a small amount of hydrochloric acid. This was incubated at room temperature, and when microscopic examination revealed the presence of yeasts or yeast-like organisms, the culture was set aside for further purification.

The second procedure employed in the isolation of the yeasts was the dilution method. A loopful of liquid containing the yeasts was transferred to a test tube containing 10 cc. of distilled water. The tube was shaken to obtain an even distribution of the organisms in the water, and then a loopful of the liquid was streaked on a plate of potato-dextrose agar or on #5 agar (1000 cc. water, 20 gms. agar, 3 gms. maltose, 1 gm. meat peptone). Two streaks were made from each culture, one plate being incubated at room temperature and one at 28 degrees Centigrade.

Growth on the plates occurred within 48 hours, the bacterial colonies usually appearing before the yeast colonies and the mold growth later than either. The individual colonies were examined microscopically, and transfers were made from yeast and yeast-like colonies to potato-dextrose broth. After incubation of the potato-dextrose broth culture for 48 hours, transfers were made to slants of potato-dextrose agar, which was used as the stock culture medium.

Repeated attempts were made to obtain sporulation in all of the cultures,

using Gorodkova's agar, carrot slants, a variation of the sporulation stock medium for yeasts and other fungi by Mrak, Phaff, and Douglas (1942) (directions for the medium were followed except that the cucumbers were omitted), and plaster of Paris blocks. In no case was sporulation obtained except with the yeasts which formed spores readily on the commonly used media. Growth on Gorodkova's agar was poor but it was good in all cases on the sporulation stock medium.

Other culture media used were: commercial beer wort², on which growth was good; liquid synthetic medium (0.1 per cent KH_2PO_4 , 0.1 per cent $(\text{NH}_4)_2\text{SO}_4$, 0.05 per cent MgSO_4 , and 5 per cent glucose), on which growth was poor or fair; and potato-dextrose broth, on which growth was good.

For testing the fermentation ability of the yeasts, potato-dextrose broth with 2 per cent of a sugar was used in large test tubes (20 mm. in diameter) with inverted Wassermann tubes for gas traps.

Twenty-five to thirty isolations were made, a number of which were of the same species. In these are included sporogenous yeasts, asporogenous yeasts, and members of the Endomycetaceae. The sporogenous yeasts and some interesting asporogenous ones, or forms in which no sporulation was obtained, are included here.

SYSTEMATIC TREATMENT AND DISCUSSION

A. Sporogenous yeasts

Hansenula sp.

Plate 31 (top, left, and bottom, right) and Plate 33, figs. 1-3

The present species was isolated from honey-locust shucks, chinaberries, and persimmons. The three cultures seemed to be identical in all characteristics studied.

Growth on potato-dextrose agar slant at first smooth or farinaceous, chalky white, becoming rugose in 48-hour cultures; in 3-day cultures folded and rose-colored; in 30-day cultures folded, avellaneous (Ridg.); vegetative cells oval and elongated with some chains of elongated cells, $2.5-4.9 \times 4.6-23$ (32.2) μ ; asci occurring singly and in chains, of same size as the vegetative cells; spores produced in 48-hour cultures, hat-shaped, 4 per ascus, averaging $2.3 \times 2.8\mu$.

Film forming on beer wort within 48 hours, smooth and chalky-white at first; in 3-day cultures folded, off-white; cells oval to very elongated or cylindrical with rounded ends, $2.5-4.9 \times 3.0-14\mu$; in older cultures, film faintly pinkish; cells from 15-day beer wort film $2.3-5 \times 3-23\mu$.

Growth on potato-dextrose broth abundant and with a film.

Film forming on liquid synthetic medium in 3 days; smooth; cells oval and elongated with some chains of elongated cells, $2.3-5 \times 6-9.2\mu$.

Fermentation of glucose and maltose. No fermentation of sucrose, galactose, or lactose.

Conjugation preceding ascospore formation was observed; however, it was seen infrequently and is apparently not usual. In the conjugating cells observed, two

² Kindly furnished in large quantity by the Anheuser-Busch Company.

cells were attached by a short, narrow conjugation tube. The male cell was small and oval with one end tapering. The female cell was two or three times as large as the male cell, spherical or ovoid, and contained several to numerous globules.

Streaks made on potato-dextrose agar slants from 90-day cultures remained white after 15 days, and very few spores were formed.

No definite conclusions were reached as to the method of ascospore formation. In some cases, it appeared that 3 cleavage furrows were formed, one horizontal furrow at either end of the cell and one vertical furrow down the remaining center section. The rims of the spores were formed along the lines of the furrows. In other cases, the spores were seen lying irregularly within the ascus. In cells with large vacuoles, a spore was formed on either end and on either side of the vacuole.

The present species does not agree well with any of the species of *Hansenula* described by Bedford (1942). Fermentation tests were not carried out for all of the sugars mentioned in Bedford's key. Since cell size varies greatly in the different media and also in the same medium at different ages and under different temperature conditions, it is difficult to use it as a basis for taxonomic determinations.

Schizosaccharomyces octosporus Beyerinck

Plate 33, figs. 4-8

A yeast which was identified as *Schizosaccharomyces octosporus* Beyerinck was collected from bark and sap from a maple tree. It grew abundantly in the potato-dextrose broth and all stages of sexual reproduction were observed. Repeated attempts to isolate it in pure culture were unsuccessful.

In both vegetative and reproductive characters the yeast resembled *Schizosaccharomyces octosporus* Beyerinck, found previously only from warm climates, except for the report of it by Coker and Wilson (1911) from Delaware grapes bought in Chapel Hill.

Guilliermond (1920) describes the genus as follows: "Round or rectangular cells, dividing by transverse partition. Asc with four or eight ascospores ordinarily resulting from isogamic copulation."

Guilliermond describes sexual reproduction in this species as follows: "The asc results from an isogamic copulation which takes place between two neighboring cells. These unite by means of a copulation canal through which the contents of the two cells mix. The fusion results in the formation of a large oval zygospore (6-10.5 μ wide and 14-20.5 μ long). This transforms slowly into an asc. Sometimes fusion remains incomplete, and the asc seems to be formed of two enlarged parts united by a canal. All intermediary stages are found, however, between complete and incomplete fusion."

Coker and Wilson found no copulation tubes or elongated processes projecting from the cells and they suggested that the copulation canal was formed by the drawing out of the cells after fusion. The present observations support their opinion.

Vegetative cells elliptical to very elongated, sometimes in chains, the individual cells of the chain bending back and remaining attached by one corner (described by Coker and Wilson, 1911) and finally breaking apart; vegetative multiplication by transverse partitioning.

Asci elongated, curved or horseshoe-shaped, or sometimes resembling two sacs connected by a canal, $3.5-4.6 \times 12.7-16.1\mu$; containing eight ascospores. Ascospores elliptical or bean-shaped, $2.7-4.1 \times 5.2-7.8\mu$.

Nematospora Phaseoli Wingard

Plate 31 (bottom, left) and Plate 33, figs. 9-13

Wingard (1922) first described *Nematospora Phaseoli* which was isolated from diseased lima beans (*Phaseolus lunatus* L.) from eastern Virginia. He has reported details of its morphology, cytology, and pathogenicity (1925).

The genus *Nematospora* is characterized by having long needle-shaped ascospores.

A culture of *Nematospora Phaseoli* Wingard, isolated in the fall of 1933 from Carolina Sieva lima beans (*Phaseolus lunatus*), was turned over to the writer by Dr. J. N. Couch.

Two transfers to potato-dextrose agar slants were made. The cultures were examined after five weeks and two types of growth were observed. Growth on slant "A", which had been folded and slightly warty in young cultures, had become smooth; while growth on slant "B" was folded and pitted and had a fringe around the edge where mycelial growth was present.

A microscopic examination of the two cultures revealed abundant asci and spores as well as small budding cells and large round cells in culture "A"; heteromorphic, small cells and few spores in culture "B". When cultures "A" and "B" were transferred to fresh slants of potato-dextrose agar, the same macroscopic and microscopic characters were retained.

Culture "A"

Growth on potato-dextrose agar slant after three days abundant, watery, off-white, convex, folded and slightly warty; in 30-day culture vegetative cells variable in size and shape, spherical, elliptical, and irregular; spherical cells $7.8-27.3\mu$; elliptical cells $4-7.8 \times 5.8-15.6\mu$. Asci and ascospores present on 48-hour potato-dextrose agar slant; asci cylindrical with rounded ends, $9.7-11.7 \times 64-88\mu$. Ascospores eight in number, in two groups of four, $3.2-4 \times 34-52\mu$; one-septate, sometimes with a bulge on the basal cell just below the septum, fusiform, with one non-motile, whip-like appendage at base.

On 55-day potato-dextrose agar slant, a few spores were seen germinating. At germination, the contents of the upper cell of the ascospore appeared to disintegrate and a swelling appeared near the upper end of the basal cell. The cell germinated by producing either bud cells or a septate pseudomycelium. This was observed also by Wingard (1925).

Culture "B"

Heteromorphic cells and pseudomycelium composed of chains of elongate cells were found in culture "B".

Wingard (1925) describes conjugation of the two ascospores as taking place by the fusion of outgrowths from their basal cells to form a conjugation canal. This phenomenon was not observed by the present writer.

Nadsonia fulvescens (Nadson and Konokotine) Sydow

Plate 31 (top, right) and Plate 32

Couch (1944) in the spring of 1943 found a yeast which he identified as *Nadsonia fulvescens* growing profusely in the sap exuding from recently cut birch stumps. In the spring of 1944, the present writer found the same yeast growing in a similar manner in the same locality. The yeast, which was found in large quantities, was again isolated in pure culture.

Members of the genus *Nadsonia* have heterogamic conjugation which takes place between a bud cell, about one-fourth the size of the parent, and the parent cell. In ascus formation, the parent cell sprouts a bud at the opposite end from the small, conjugating bud cell. The fused contents of the parent and the small bud cell pass into the ascus. A single spiny-walled spore is formed.

The yeast was forming spores abundantly when it was collected in the sap of the birch tree. It also produced asci abundantly on potato-dextrose agar.

Growth on potato-dextrose agar slant abundant, smooth, raised; light cream-colored at first, turning brown when spores are produced; cells oval, elliptical and elongate-apiculate, $5.8-8 \times 9.2-18.4\mu$, occurring singly, in twos, or in chains of three, budding at one or both ends. Asci produced abundantly on potato-dextrose agar, oval, with end attached to parent cell truncated, $5-8 \times 5.8-11.5\mu$, containing a single spore, or infrequently two spores which are not completely separated. Ascospores yellowish-brown, spherical, minutely spiny-walled, containing one or sometimes two large oil globules, $4.6-6.9\mu$. Couch (1944) found elongated asci which measured up to 10.5μ in width and 16μ in length.

Growth on beer wort without a film, abundant, and with the formation of sediment.

B. Asporogenous yeasts

For the identification of the asporogenous yeasts, parts of Lodder's key (1934), which are given by Henrieci (1941), were used. Generic determinations are tentative and specific identifications were not attempted. Although numerous attempts were made to induce sporulation in the following forms, none occurred; however it cannot be concluded that these are necessarily asporogenous yeasts. Under proper conditions of temperature, moisture, and food supply, sporulation might occur.

Rhodotorula sp.

Plate 33, fig. 14

Lodder includes all of the red yeasts in the family Rhodotorulaceae and the 13 species and 10 varieties in this family are included in the single genus *Rhodotorula*. The present species was isolated from privet (*Ligustrum*), dogwood (*Cornus*), and barberry (*Berberis*) fruits.

Growth on 3-day potato-dextrose agar slant abundant, smooth, glistening, convex, strawberry pink (Ridg.); periphery of streak even; cells spherical, oval or ellipsoid, $2.3-4.6 \times 2.8-7.1\mu$, containing one or two large oil globules; budding at one end only.

Growth in 10-day beer wort culture without a film but with a faint ring; cells containing many small oil globules; in 14-day beer wort cultures a thick, mucoid, pinkish film present.

Kloeckera sp.

Plate 34, fig. 1

The genus *Kloeckera* includes those asporogenous yeasts which are mostly lemon-shaped and have bipolar budding. The present species was isolated from *Elaeagnus* and apple (*Malus*) fruits.

Growth on 3-day potato-dextrose agar slant smooth, shiny, off-white, spreading; margin of streak wavy; cells mostly apiculate, some oval and elongated, $2.3-5.3 \times 2.8-9.2\mu$; budding bipolar.

Growth in 3-day beer wort culture without a film, abundant, and with the formation of sediment.

Vigorous fermentation of glucose.

Mycoderma sp.

Plate 34, figs. 2, 3

This genus is characterized by the formation of pseudomycelium of elongated cells and by the formation of a dry pellicle in wort cultures. The present species was isolated from a jelly fungus, *Eridia gelatinosa*. A distinct alcoholic odor was formed when the yeast was growing on the fungus in potato-dextrose broth.

Growth on 5-day potato-dextrose agar slant abundant, off-white, dull, finely warty; margin on slant irregular; cells oval, elongate and irregularly shaped, $2.3-6.9 \times 3.5-18.9\mu$; budding multipolar; pseudomycelium formed; on 20-day potato-dextrose agar slant, growth much folded and convoluted; light buff in color.

On beer wort, scum forms quickly; thick, light buff, and folded in 10-day cultures; composed of chains of elongated cells forming a pseudomycelium.

On sporulation stock medium, growth is abundant, raised, and much convoluted.

Vigorous fermentation of glucose.

Torulopsis sp. "A"

Plate 34, figs. 4-6

The members of this genus are characterized by having oval or cylindrical cells and no pellicle in wort cultures. The present species formed long, slender tubular processes resembling copulation tubes on Gorodkova's agar, on the sporulation stock medium, and on potato-dextrose agar. Asporogenous forms which produce processes resembling copulation tubes are put by Lodder in the genus *Asporomyces*. Mrak, Phaff, and Smith (1942) consider the genus *Asporo-*

myces non-valid because other imperfect yeasts also produce such processes. Isolations of the present species were made from hawthorn apples (*Crataegus*), from soil and persimmon seeds from underneath a persimmon tree, and from rose apples (*Rosa*).

Growth on 5-day potato-dextrose agar slant smooth, slightly shiny, spreading, pinkish buff (Ridg.); periphery of streak wavy; cells oval, ellipsoid or elongated, rounded or tapering at the ends, $2.3-4.6 \times 3-11.5\mu$; budding from one or both ends; on 21-day potato-dextrose agar slant some heteromorphic cells present, which have tubular outgrowths somewhat resembling copulation tubes.

Growth on 3-day beer wort culture abundant and with the formation of sediment.

No fermentation of glucose.

Torulopsis sp. "B"

Plate 34, fig. 8

The present species was isolated from fruits of yaupon (*Ilex vomitoria*).

Growth on 10-day potato-dextrose agar slant abundant, raised, shiny, very light buff; cells oval or ellipsoid, $4.1-7.4 \times 4.6-10.4\mu$, usually containing one or several large globules.

Growth in 14-day beer wort culture without a film and with a slight sediment.

Growth on 50-day sporulation stock medium dull, smooth, light buff.

Fermentation of glucose vigorous.

Torulopsis sp. "C"

Plate 34, fig. 9

Growth on 3-day potato-dextrose agar slant smooth, glistening, raised, light cream colored or off-white; periphery of streak even; cells spherical or oval, $2.5-4.6 \times 2.5-6.9\mu$; budding multipolar.

Growth in 10-day beer wort culture abundant and with a faint film.

Vigorous fermentation of dextrose.

Trigonopsis sp.

Plate 34, fig. 7

Triangular cells, budding at the three angles, serve to characterize this genus. The present species was isolated from a fungus, *Sebacina* sp., which was growing on dead wood.

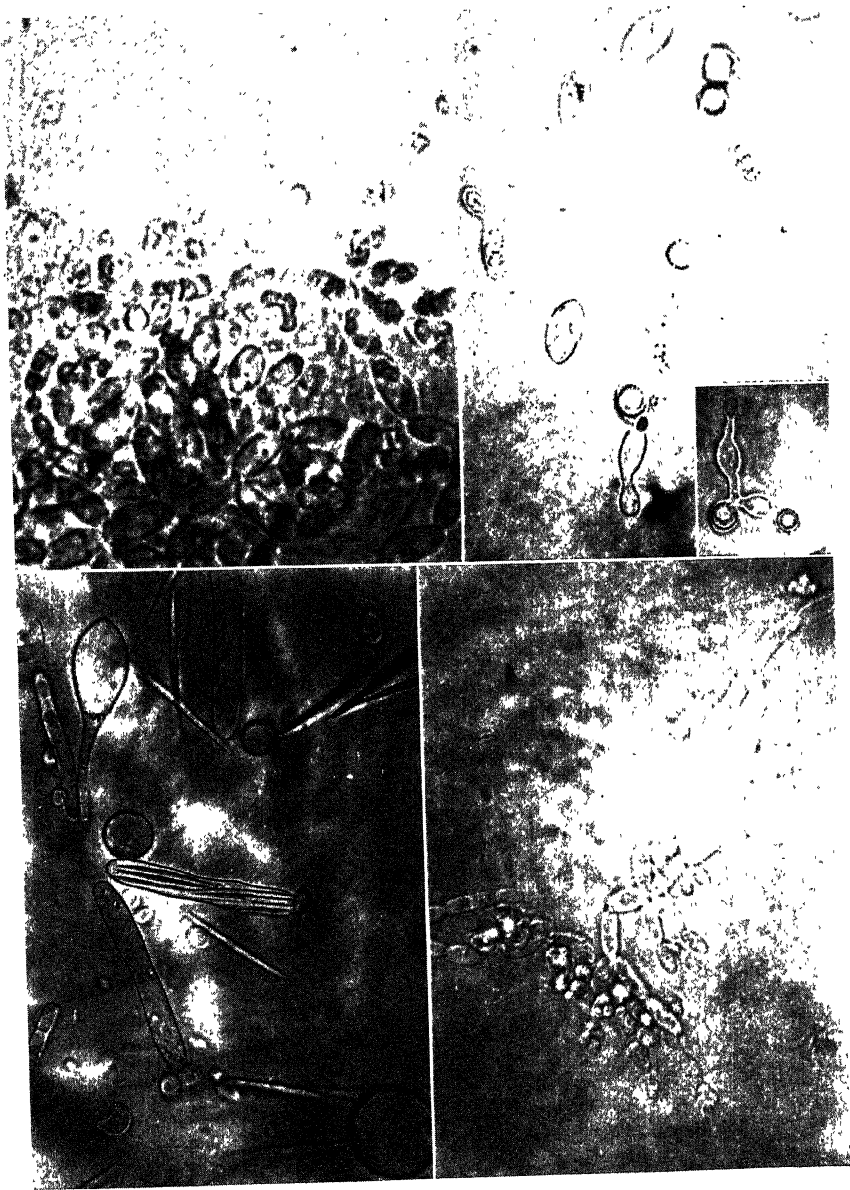
Growth on 3-day potato-dextrose agar slant off-white, smooth, dull or only slightly shiny; mycelial growth along edges of streak; cells mostly triangular with rounded angles, $1.8-4.6 \times 2.8-15\mu$; pseudomycelium composed of elongated, branching cells.

In 10-day beer wort cultures a faint film is formed; no mycelium or pseudomycelium, the cells occurring singly or in groups of a few budding cells.

SUMMARY

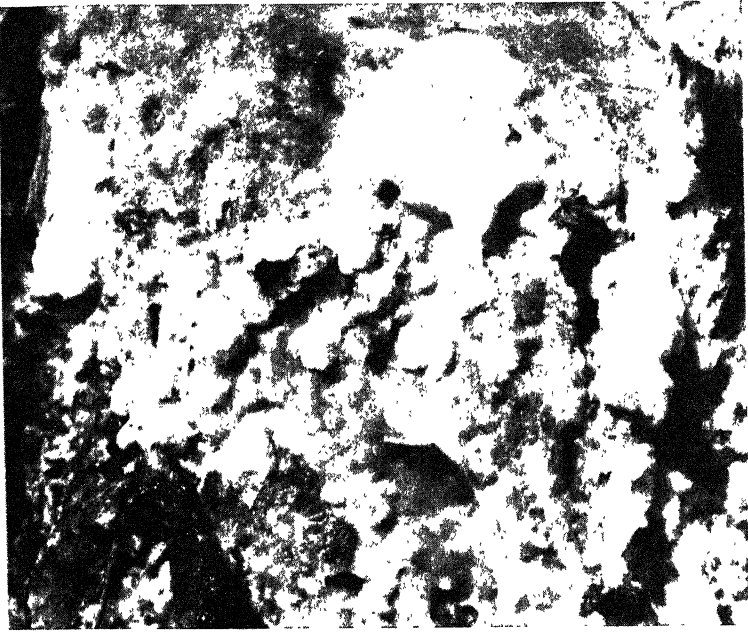
Collections were made of wild fruits, bark, wood, and sap of trees, a few fungi growing on dead wood, and of soil in the vicinity of Chapel Hill. Yeasts were

PLATE 31



Hansenula sp. Photomicrograph of asci and ascospores (top, left), $\times 958$; chains of vegetative cells and asci (bottom, right), $\times 640$.
Nadsonia fulvescens. Photomicrograph of vegetative cells and asci (top, right) and (inset) empty copulating cells and asci, $\times 958$.
Nematospora Phaseoli. Photomicrograph of vegetative cells and asci (bottom, left) $\times 640$.

PLATE 32



Nadsonia fulvescens. Growth on birch stump, showing a close-up view at top.

PLATE 33

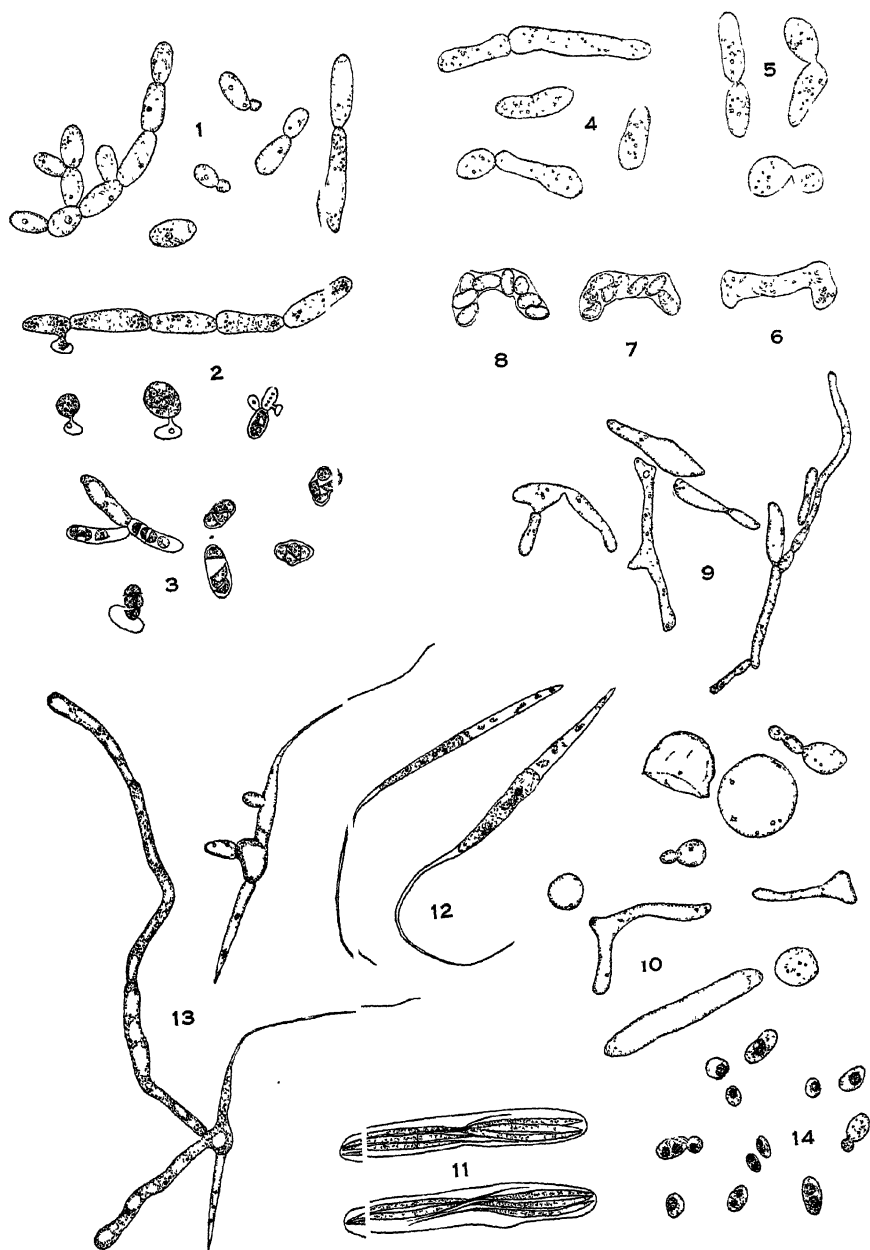


PLATE 34



isolated from a number of collections. Eleven wild yeasts were studied and were identified in so far as possible. A culture of *Nematospora Phaseoli* Wingard, isolated from lima beans by Dr. J. N. Couch, was included in the study. The following spore-forming yeasts are described in this paper: *Hansenula* sp., *Schizosaccharomyces octosporus* Beyerinck, *Nematospora Phaseoli* Wingard, and *Nadsonia fulvescens* (Nadson and Konokotine) Sydow. Asporogenous forms described are: *Rhodotorula* sp., *Kloeckera* sp., *Mycoderma* sp., *Torulopsis* sp. "A", *Torulopsis* sp. "B", *Torulopsis* sp. "C", and *Trigonopsis* sp. These forms have been illustrated by photomicrographs and drawings.

ACKNOWLEDGMENTS

The writer wishes to express her sincere appreciation to Dr. J. N. Couch for the assistance given and interest shown during the course of this study, and to Mrs. C. D. Beers for arranging plate figures.

LITERATURE CITED

- ANDERSON, K. W., AND C. E. SKINNER. 1947. Yeasts in decomposing fleshy fungi. *Mycologia* **39**: 165-169.
- BEDFORD, C. L. 1942. A taxonomic study of the genus *Hansenula*. *Mycologia* **34**: 628-649.
- COKER, W. C., AND LOUISE WILSON. 1911. *Schizosaccharomyces octosporus*. *Mycologia* **3**: 283-287.
- CONANT, NORMAN F. 1940. The taxonomy of the anasco-sporous yeast-like fungi. *Mycopathologia* **2**: 253-266.
- COUCH, J. N. 1944. The yeast *Nadsonia* in America. *Jour. Elisha Mitch. Sci. Soc.* **60**: 11-16.
- GUILLIERMOND, A. 1920. The Yeasts. Translated by F. W. Tanner. John Wiley and Sons, New York.
- HENRICI, ARTHUR T. 1941. The yeasts. *Bacteriological Reviews* **5**: 97-179.
- KURTH, E. F. 1946. Yeasts from wood sugar stillage. *Industr. Engng. Chem.* **38**: 204-207.
- KURTH, E. F., AND V. H. CHELDELIN. 1946. Feeding yeasts from wood sugar stillage. *Industr. Engng. Chem.* **38**: 617-619.
- LEHMAN, S. G. 1943. Occurrence of yeast spot on soybean in North Carolina. *Plant Dis. Reporter* **27**: 602.
- LODDER, J. 1934. Die Hefesammlung des "Centraalbureau voor Schimmelcultures." II Teil. Die Anaskosporogenen Hefen. Erster Halfte. Amsterdam.
- MRAK, E. M. 1945. Microbiological deterioration in dried and dehydrated fruits. *Canner*, **c**, **17**: 22.
- MRAK, E. M., AND L. S. McCLUNG. 1940. Yeasts occurring on grapes and in grape products in California. *Jour. Bacteriol.* **40**: 395-407.
- MRAK, E. M., H. J. PHAFF, AND H. C. DOUGLAS. 1942. A sporulation stock medium for yeasts and other fungi. *Science* **96**: 432.
- MRAK, E. M., H. J. PHAFF, AND B. L. SMITH. 1942. Non-validity of the genus *Asporomyces*. *Mycologia* **34**: 139-141.
- NICKERSON, W. J., JR. 1943. *Zygosaccharomyces acidifaciens*: a new acetifying yeast. *Mycologia* **35**: 66-78.
- . 1944. Studies on film-forming yeasts. Acid production by *Zygopichia* and *Zygo-hansenula*. *Mycologia* **36**: 224-233.
- RIDGWAY, R. 1912. Color Standards and Color Nomenclature. Washington, D. C.
- WINGARD, S. A. 1922. Yeast-spot of lima beans. *Phytopath.* **12**: 525-532.
- . 1925. Studies on the pathogenicity, morphology, and cytology of *Nematospora Phaseoli*. *Bull. Torr. Bot. Club* **53**: 249-290.

PLATE 33

Hansenula sp.

Fig. 1. Vegetative cells from potato-dextrose agar culture, $\times 850$.

Fig. 2. Conjugating cells from 3 and 6-day potato-dextrose agar cultures, $\times 850$.

Fig. 3. Asci and ascospores from potato-dextrose agar culture, $\times 850$.

Schizosaccharomyces octosporus Beyerinck

Fig. 4. Vegetative cells. One cell dividing by transverse partitioning, $\times 850$.

Figs. 5-8. Stages in fusion and formation of ascus, $\times 850$.

Nematospora Phaseoli Wingard

Fig. 9. Heteromorphic cells from culture "A" on potato-dextrose agar, $\times 525$.

Fig. 10. Vegetative cells from culture "B" on potato-dextrose agar, $\times 525$.

Fig. 11. Asci containing ascospores, $\times 525$.

Fig. 12. Ascospores, $\times 850$.

Fig. 13. Germinating ascospores, $\times 850$.

Rhodotorula sp.

Fig. 14. Vegetative cells from 3-day potato-dextrose agar culture, $\times 850$.

PLATE 34

Kloeckera sp.

Fig. 1. Vegetative cells from 3-day potato-dextrose agar culture, $\times 850$.

Mycoderma sp.

Fig. 2. Pseudomycelium from film of 10-day beer wort culture, $\times 850$.

Fig. 3. Budding cells from 3-day potato-dextrose agar culture, $\times 850$.

Torulopsis sp. "A"

Fig. 4. Vegetative cells from 3-day potato-dextrose agar culture, $\times 850$.

Fig. 5. Vegetative cells from 21-day potato-dextrose agar culture showing tubular processes resembling copulation tubes, $\times 850$.

Fig. 6. Vegetative cells from Gorodkova's agar culture showing tubular processes resembling copulation tubes, $\times 850$.

Trigonopsis sp.

Fig. 7. Vegetative cells from 3-day potato-dextrose agar culture, $\times 850$.

Torulopsis sp. "B"

Fig. 8. Vegetative cells from 10-day potato-dextrose agar culture, $\times 850$.

Torulopsis sp. "C"

Fig. 9. Vegetative cells from 3-day potato-dextrose agar culture, $\times 850$.

KERATINOPHILIC PHYCOMYCETES

I. A NEW GENUS OF THE SAPROLEGNIACEAE¹

BY MAEBURN BRUCE HUNEYCUTT

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina*

PLATES 35 AND 36

INTRODUCTION

Perhaps the occurrence of soil and water fungi on animal material such as chitin and keratin-containing tissues is much more extensive and common than is generally realized. Petersen (1903) in Denmark was the first to study these saprophytes occurring on chitinous materials. These fungi were neglected in the United States until 1937, when Sparrow made an extensive study in which he recorded Petersen's Danish species and described several new American species occurring on the exuviae of various insects. Karling (1945) reported several additional species from the Amazon Valley in Brazil, thus indicating that chitinophilic fungi are widespread and possibly rather common throughout the world. These fungi were called chitinophilic because they are chitin-loving.

Karling in 1946 introduced the use of keratinized tissues of animals for isolating and growing chytrids. Several new species were discovered and described (1946, 1947). He found human skin to be the most favorable keratinized substratum. However, the "down" of ducks and geese, pinnae of other feathers, and fine shreds of wool as well as hair of babies and other immature animals were also found to be favorable substrata. He reported that shavings of nails, hoofs, and horns were too thick and hard for use in microscopic studies although they provided nourishment.

METHODS OF COLLECTION AND CULTURE

In this study, although particular emphasis has been placed on the lower forms of aquatic and soil Phycomycetes, the higher ones have not been overlooked. In the first part of the study, emphasis was placed chiefly on those forms saprophytic on vegetable matter for the most part. Recently, emphasis has shifted to those forms occurring chiefly on keratin-containing animal tissues. Most of the collections have been of soil, with very few water collections being made. The greater number of the soil samples have been taken from the edges of lakes, streams, and from boggy areas which are flooded part of the year. However, several collections have been made from dry places such as plowed fields and upland waste places.

¹ This was a thesis submitted to the Faculty of the University of North Carolina in partial fulfillment of the requirements for the degree of Master of Arts in the Department of Botany.

The author's present address is University of Tennessee, School of Pharmacy, Memphis 3, Tenn.

Following the work of Karling and others, considerable interest was developed in those forms growing on chitinous and keratin-containing materials, particularly the latter. Therefore, in making collections the possibility has not been overlooked that if one collected soil from places where keratin-containing materials naturally occur, keratinophilic fungi might also occur. Hence, recently collections have been made with this in mind. Several collections have been made around old blacksmith shops where horses and mules have been shod and pieces of hoof and fallen hair have been lying for many years. Collections have also been made from old pastures where hair has dropped from cattle and from old hog pastures and places where hogs have been butchered periodically, the hair being left to decay and perhaps attacked by keratinophilic fungi. Results have not been disappointing.

The method of collecting was the usual one employed in this laboratory. Small amounts of soil, about two tablespoonfuls, were collected in small wide-mouth bottles. These were poured separately into sterile petri dishes and enough sterile charcoal water added to cover the soil completely. Then various materials were added as "bait." Among the substrata used have been leaves of various grasses, especially *paspalum* and corn, small pieces of cellophane, filter paper, human hair, especially red hair, human skin, shavings of horse hoof, cow horn, and bits of shrimp exoskeleton. All these materials have been sterilized by boiling before use. Very thin shavings of horse hoof have been found satisfactory as a substratum upon which to isolate certain chytrids and other keratinophilic members of the lower *Phycomycetes*. Human hair has been very satisfactory and some species have been isolated on it.

Another technique used was that of drying the soil for several days, then wetting again with sterile charcoal water and adding various substrata. This was done at first in order to isolate members of the *Blastocladales*. Although little emphasized, this technique was used by Couch and Whiffen in 1942 in isolating species of *Blastocladiella*.

The present study concerns primarily a member of the *Saprolegniaceae* which occurred in a soil sample collected in a pasture near Chapel Hill, North Carolina. The soil was brought into the laboratory in October, 1947. Several materials were added at that time as substrata, including shavings of horse hoof. After a perfunctory check, nothing of great interest was found. In the spring of 1948, it was allowed to dry several days along with many other collections which had been in the laboratory for several weeks. Sterile charcoal water was added again and "baited" this time with pieces of horse hoof and *paspalum* grass leaves. In the process of examining some pieces of hoof after several days, an interesting fungus looking much like *Aphanomyces* was particularly noted. It was peculiar because the oogonia generally contained more than one egg each, which is different from *Aphanomyces*. Upon closer examination, the encysted spores were observed to be enclosed in a sporangium.

Structurally, this fungus is characterized by delicate, much-branched hyphae, predominantly globose to subglobose sporangia with the spores always encysting within the sporangium, and papillate oogonia usually containing more than one very thick-walled egg each.

Thus the fungus appears distinct from any other genus in the Saprolegniaceae. It is diagnosed as a new genus and described below.

DESCRIPTION OF GENUS AND SPECIES

Aphanodictyon gen. nov.

Mycelium of limited growth, hyphae very slender as in the more delicate species of *Aphanomyces*, much branched; sporangia variable in shape but predominantly globose to subglobose; spores encysting within the sporangium, emerging separately from their cysts as in *Dictyuchus*, leaving a net of empty cysts, and swimming in the laterally biflagellate form or germinating *in situ* by germ tube. Oogonia abundant, containing from one to several thick-walled eggs. All mature cell walls give a cellulose reaction with chloroiodide of zinc.

Aphanodictyon papillatum sp. nov.

Mycelium of limited growth on horse hoof, hyphae delicate, up to 6 mm. long, usually less, about 1-6 μ thick, usually 3-5 μ , much branched and usually with many short lateral branches; sporangia borne terminally on main and lateral hyphae, sporangial stalk small, usually about 2 μ thick, variable in shape but predominantly globose and subglobose, up to 23 x 33 μ , varying frequently to ellipsoid, obovate, oblong or kidney-shaped, about 7-26 x 12-44 μ ; spores one to many, encysting within the sporangium, emerging separately from their cysts as in *Dictyuchus*, leaving a net of cyst walls, and swimming in laterally biflagellate form, or germinating *in situ* by a germ tube, sharply polygonal when first differentiated, later rounding up but with the sides in contact, usually remaining somewhat flattened, globose to subglobose, 6-12 x 7-15 μ , sometimes elongate, up to 8 x 24 μ , or irregular in shape; oogonia developed abundantly, terminal on main and lateral hyphae or intercalary, very variable in shape, globose to subglobose, 14-35 x 15-39 μ , ellipsoid, pyriform, oblong to cylindric with rounded ends, about 19-32 x 27-59 μ , infrequently roughly rectangular in outline, up to 27 x 44 μ or very irregular and up to 31 x 49 μ , eggs one to seven in an oogonium, usually subglobose to ellipsoid, rarely perfectly globose, about 12.5-20 x 14-24 μ , sometimes triangular in outline with rounded ends, the sides in contact somewhat flattened; egg walls very thick and variable in thickness in the same egg, about 1-5 μ thick; refractive globule usually slightly excentric, sometimes centric, about 6.8-9.3 μ in diameter; papillae on oogonia few to numerous, variable in length, up to 20 μ , usually about 2 μ wide; antheridia always present, androgynous or diclinous.

Saprophytic on keratinized materials, in pasture near Chapel Hill, North Carolina.

Since the fungus has grown best on horse hoof, all study has been on living material growing on thin shavings of hoof. When detailed observations were first begun, it was attempted to dissect out the hyphae bearing the sporangia or oogonia that were desired. This was soon found to be unsatisfactory. In the first place, if the fine steel needles which were used in dissecting were slightly rough the delicate hyphae clung tenaciously to them and usually could not be disengaged without considerable mutilation and injury. It was found that if pieces were small enough, whole mounts could be very successfully made. First the fungus was allowed to grow about three days on a large but thin piece of hoof. When it had become well covered with the mycelium, the large piece was

cut into small ones not over 2 sq. mm. These were placed in sterile petri dishes and washed. In a few days sporangia and oogonia appeared. These pieces, when the fungus was in the desired stage of development, were floated on a clean slide in order not to entangle the hyphae. For detailed structure, number zero or number one cover glasses were used. Observations were made with a 70 \times apochromat water immersion objective at first, later switching to an apochromat 2 mm. oil objective with N. A. of 1.4. Most of the drawings were made with the latter objective.

Since *Aphanodictyon* grows very well on shavings of horse hoof and since this is a rather selective medium, it was relatively easily gotten into unifungal culture. Two or three methods were used. Perhaps the easiest way was by transferring single sporangia to sterile pieces of hoof. Single sporangia were isolated in this way: A small bit of mycelium bearing several mature sporangia was transferred to a sterile 2% agar plate. The sporangia are relatively large and could be observed under an ordinary binocular dissecting microscope. With a fine-pointed needle a single sporangium was pushed away from the entangling hyphae. A small block of agar was then cut out around the sporangium and transferred to a shaving of hoof in a sterile petri dish. The spores germinated readily.

Preliminary attempts have been made to get this fungus into absolutely pure culture on agar. It was first tried on 2.5% plain agar. Single sporangia were pushed about on an agar plate in order partially to free them from bacteria. These were then transferred on small blocks of agar to sterile 2.5% agar plates. The spores germinated, sending out long germ tubes, but these soon stopped growing. Several attempts have been made to get it to grow on agar by transferring bits of mycelium to various agars containing different amounts of peptones. Agars were used containing .01%, 0.15%, and 1% peptone as well as agar to which powdered hoof of various amounts had been added. All of these attempts failed.

Although no extensive experiments have been carried out to determine the range of substrata on which *Aphanodictyon* will grow, it has been grown successfully on human skin, human hair, shavings of cow horn, shavings of horse hoof and pieces of hemp seed. Up to the present time it has failed to grow on shrimp exoskeleton and on pieces of boiled paspalum grass leaves.

DEVELOPMENT OF SPORANGIA AND OOGONIA

In the very early stages of growth, the hyphae look very much like *Aphanomyces*. They are very delicate and slender, becoming much branched after a few days of growth. The mycelium forms a delicate translucent halo around pieces of hoof, usually extending 3 mm. out into the water after approximately two weeks of growth.

Although not enough data have been accumulated nor conditions of growth sufficiently standardized to make general statements as to the time of development of sporangia, in those cultures which have been timed, sporangia have been noted on the fifth or sixth day after inoculation. Sporangia are developed

abundantly. They arise as globose swellings at the ends of main and lateral hyphae. A main hypha may have several branches which bear sporangia. The main hyphae may or may not bear sporangia at their tips; if not, they apparently continue to grow in length. Very early stages of development have not been followed, but a central vacuole was present in later stages in all those observed closely. As the sporangia grow, the protoplasm becomes dark, appearing densely granular. The central vacuole changes shape and becomes larger. Irregular cleavage furrows can be seen extending toward the sporangial wall. These furrows ultimately cut the sporangial protoplasm into a few to many spore initials. A disappearance stage then occurs in which the spores seem to coalesce and the spore limits become indistinct. When the spores again become differentiated, they are polygonal from pressure. They later become rounded, but the sides in contact usually remain somewhat flattened. The cleavage of spores by furrows seems to be essentially the same as that described for *Saprolegnia* and *Achlya* by Humphrey (1892), Trow (1899), and others. However, one point in development is very puzzling. It is not possible to state whether or not a cross wall is formed at the base of the sporangium as described for other members of the Saprolegniaceae. In some instances, an apparent membrane could be seen, but it usually appeared in the sporangial stalk, some distance below the sporangium. The nature of this membrane is not known. In other instances no membrane could be seen. In some sporangia, a spore may become narrow and extend into the sporangial stalk.

The number of spores in the sporangia varies greatly. Several sporangia have been seen with only one spore, many with two to several, but usually there are several dozen spores in each sporangium.

The sporangia vary greatly in size and shape. However, the globose and subglobose ones are predominant. They vary to ovate, pyriform, ellipsoid and oblong with rounded ends. Infrequently they are kidney-shaped. The globose to subglobose sporangia measure up to $22.5 \times 33\mu$. In the ellipsoid-ovate-oblong group, the largest found measured $26 \times 44\mu$. The largest kidney-shaped sporangium measured $22 \times 44\mu$.

In the first part of the study, zoospores were not seen to emerge, although the spores had been seen to germinate by germ tube within the sporangia. The first zoospores which were seen to emerge were in a water mount which had been made several hours. It was found that zoospores could be induced to swim by washing and adding fresh, sterile, charcoal water. The first stage noticeable in the emergence of the zoospores is a slight bulging of the spore and sporangial walls at the point where the spore is going to emerge. A more or less circular hole is digested in the spore and sporangial walls and the spore flows slowly out, assuming at first a rounded shape. As it slowly differentiates into the kidney-shaped spore, the flagella can first be seen growing out from the spore as short bristles. They wave slowly at first, the free ends moving in a circular path. As they grow in length, the rate of movement increases. Until fully differentiated, the zoospore remains apparently attached near the pore through which it emerged. Just before the zoospore swims, the flagella stop moving and the zoospore appears

to be pulling to get free, becoming a little distorted. Suddenly, it becomes detached and glides away in a smooth, spiral movement. The above observations on the emergence of the zoospores from the cysts agree with those made by other observers on the Saprolegniaceae. Whether the zoospores repeatedly encyst and emerge as was described for those of *Dictyuchus* by Weston (1919) has not been determined.

The oogonia are developed abundantly, usually appearing after the sporangia. They arise as terminal swellings on the main hyphae or on lateral branches or less frequently as intercalary swellings. The oogonia very early become papillate. Since no early stages have been followed for any period of time, it cannot be said when a central vacuole is formed. The vacuole is present in all cases in all those oogonia observed approaching mature size. Although several attempts have been made to follow the stages just preceding and including egg formation, this has not been achieved. It could be followed without a cover glass with a $21\times$ objective, but due to the small size of the oogonia and the densely granular protoplasm, those observations proved unsatisfactory. Development was stopped by adding a cover glass and hence could not be followed with a water immersion objective. Enough stages have been seen, however, to be reasonably sure that development is essentially the same as occurs in the other members of the Saprolegniaceae. It was impossible to determine whether or not a cross wall is formed. This problem as well as the nuclear story must await a detailed study of the development and cytology of this fungus.

The oogonia vary greatly in size and shape. Sometimes they are globose. The most common shape is globose to subglobose. These vary from 14×17 to $34 \times 35\mu$. Frequently, they vary to ellipsoid up to $32 \times 47\mu$. Sometimes they are more or less oblong with two eggs in linear series to elongate-cylindric with four eggs in a single series. The largest oblong oogonium measured was $23 \times 59\mu$. A very few are roughly rectangular in outline and up to $27 \times 44\mu$. Some are irregular in shape, sometimes bulging and irregularly lobed. The largest of these measured $31 \times 49\mu$.

The papillae on the oogonia vary greatly from mere knobs to elongate and slender. The longest measured was about 20μ long. They are usually about 2μ thick.

The number of eggs in an oogonium varies from one to seven. In one instance it was thought that eight could be seen in a very large oogonium. This point could not be settled. More than seven eggs probably occur in an oogonium in rare instances. Oogonia containing one egg are not uncommon. Those containing two, three, and four eggs are very common. Several have been observed with five and six eggs. Only a few have been seen with seven.

The eggs vary in size and shape with the subglobose ones predominant. Globose eggs are not common. Those that are ellipsoid are quite frequently seen. The eggs almost fill the cavity of the oogonium. Usually the sides in contact are flattened from pressure. An exceptionally thick wall is formed. It usually varies in thickness in the same egg. The thickest wall measured was 5μ .

A refractive globule is formed which is usually slightly excentric, some having been seen that were centric.

Antheridia arise as slender branches from the oogonial stalk or from another hypha. Thus, they are either androgynous or diclinous. The antheridial stalks may branch and in some instances the antheridium appears branched and irregular. It is not possible to state whether or not a cross wall cuts off an antheridium.

DISCUSSION

It is very interesting to consider the possible relationships of this peculiar fungus. In considering relationships in the Saprolegniaceae several characters must be taken into account, among the most important of which are: mycelium, type of growth and extent, size, and appearance of hyphae, size and shape of sporangium, formation of spores, spore discharge and type of spore, type of oogonium and number and structure of eggs.

Aphanodictyon bears a very striking resemblance to *Aphanomyces* in the appearance and size of the hyphae. In *Aphanomyces* the hyphae are slender and delicate and sparingly to much branched. It is not possible to distinguish *Aphanodictyon* hyphae from *Aphanomyces* before sporangia or oogonia are formed. The hyphae of *Aphanodictyon* are slightly smaller than those of most species of *Aphanomyces*. However striking may be the resemblance of mycelial characters, the sporangia and oogonia are distinctly different. Sporangia in *Aphanomyces* are formed from unchanged hyphae, the spores emerging apically and encysting there in a clump. In *Aphanodictyon*, the sporangia arise as terminal swellings, and spores encysting within the sporangium.

The oogonia of *Aphanomyces* contain only one egg each, those of *Aphanodictyon* usually contain more than one. The eggs of *Aphanodictyon* have exceptionally thick walls, the walls being thicker in some places than others.

The manner of spore encystment and separate emergence in *Aphanodictyon* is very similar to that in *Dictyuchus*. When all spores have emerged, a net of empty cysts remains within the sporangium. The sporangia of the two genera are quite different in shape. Those of *Dictyuchus* are cylindrical, those of *Aphanodictyon* usually globose. The hyphae of *Dictyuchus* are much larger than those of *Aphanodictyon*. Also, oogonial characters are quite different. The oogonia of *Dictyuchus* have only one egg each.

In the Saprolegniaceae (Coker, 1923; Coker and Matthews, 1937), besides *Dictyuchus*, there are five other genera in which the spores regularly encyst within the sporangium. These are: *Aplanes*, *Thraustotheca*, *Calyptralegnia*, *Brevilegnia*, and *Geolegnia*. The members of these generally differ so greatly from *Aphanodictyon* in hyphal, sporangial, oogonial and egg characters that it is not considered worthwhile to make a detailed comparison of *Aphanodictyon* with them.

Thus on the basis of similarities in size and appearance of the hyphae, it is believed that *Aphanodictyon* is more closely related to *Aphanomyces* than to any of the other genera in the Saprolegniaceae.

On the basis of sporangial shape, size of the delicate hyphae, and the number of exceptionally thick-walled eggs to an oogonium, *Aphanodictyon* is distinct from any other genus in the Saprolegniaceae.

SUMMARY

A new genus of the Saprolegniaceae is described. It is characterized by delicate, much-branched hyphae, predominantly globose to subglobose sporangia with spores always encysting within the sporangium, emerging and swimming as in *Dictyuchus* or germinating by germ tubes; and papillate oogonia which usually contain more than one exceptionally thick-walled egg each, and its growth on keratin-containing tissues. The newly described fungus is named *Aphanodictyon papillatum*. On the basis of similarities of mycelial characters, it is probably more closely related to *Aphanomyces* than to any other genus of the Saprolegniaceae.

ACKNOWLEDGMENTS

The writer wishes to express his sincere gratitude to Dr. J. N. Couch for his constant interest, guidance, and patience throughout this work.

For suggestions pertaining to descriptions and for aid in arranging plates he is grateful to Mrs. Alma H. Beers.

BIBLIOGRAPHY

- ANTIKAJIAN, GRACE. 1947. *Rhizophidium chitinophilum*. Mycologia 39: 612-616.
- COKER, W. C. 1923. The Saprolegniaceae, with Notes on Other Water Molds. University of North Carolina Press, Chapel Hill, N. C.
- COKER, W. C., AND VELMA D. MATTHEWS. 1937. Saprolegniales (Saprolegniaceae, Ectrogellaceae, Leptomitaceae). North American Flora 2: Pt. 1. The New York Botanical Garden, New York City.
- COUCH, J. N. 1926. Notes on the Genus *Aphanomyces*, with a description of a new semi-parasitic species. Journ. Elisha Mitch. Sci. Soc. 41: 213-226.
- . 1939. Technique for collection, isolation, and culture of chytrids. Jour. Elisha Mitch. Sci. Soc. 55: 208-214.
- AND A. J. WHIFFEN. 1945. Observations on the Genus *Blastocladiella*. Amer. Journ. Bot. 29: 582-591.
- HUMPHREY, J. E. 1892. The Saprolegniaceae of the United States. Trans. Amer. Phil. Soc. 17: 63-148.
- KARLING, J. S. 1945. Brazilian chytrids. VI. *Rhopalophlyctis* and *Chytriomycetes*, two new chitinophilic operculate genera. Amer. Journ. Bot. 33: 362-369.
- . 1946. Keratinophilic chytrids. I. *Rhizophidium keratinophilum* n. sp., a saprophyte isolated from human hair, and its parasite, *Phlyctidium mycetophagum* n. sp. Amer. Journ. Bot. 33: 751-757.
- . 1947. Keratinophilic chytrids. II. *Phlyctorchiza variabilis* n. sp. Amer. Journ. Bot. 34: 27-32.
- . 1947. *Lagenidium humanum*, a saprophyte isolated on dead human skin. Mycologia 39: 224-230.
- PETERSEN, H. E. 1903. Note sur les Phycomycètes observés dans les téguments vides des nymphes de Phryganées avec description de trois espèces nouvelles de Chytridinées. Journal de Botanique 17: 214-222.
- SPARROW, F. K., JR. 1937. Some chytridiaceous inhabitants of submerged insect exuviae. Proc. Amer. Phil. Soc. 78: 23-53.

- 1943. Aquatic Phycomycetes Exclusive of the Saprolegniaceae and Pythium. The University of Michigan Press, Ann Arbor, Mich.
- Trow, A. H. 1899. Observations on the biology and cytology of a new variety of *Achlya americana*. Ann. Bot. **13**: 131-179.
- Weston, W. H., Jr. 1919. Repeated zoospore emergence in *Dictyuchus*. Bot. Gaz. **68**: 287-296.

EXPLANATION OF PLATES

Drawings made with the aid of Spencer camera lucida. All figures $\times 1035$, with the exception of figures 1, 8, 19, and 21.

PLATE 35

- Fig. 1. Habit sketch showing branched hyphae, sporangia and oogonia. $\times 235$.
- Figs. 2-7. Sporangia with two to many spores.
- Fig. 8. Sporangium showing polygonal spores before becoming rounded. $\times 813$.
- Figs. 9, 10. Ellipsoid and obovate sporangia respectively.
- Figs. 11, 12. Stages in emergence and differentiation of zoospores, having a net of empty cysts within the sporangia.
- Fig. 13. Sporangium containing one spore and an empty spore cyst.

PLATE 36

- Figs. 14-18. Oogonia of various sizes and shapes with one to several mature eggs. Thick egg wall shown by concentric dashes. Note uneven thickness of egg wall. The slightly eccentric globule is shown in lighter stippling.
- Fig. 19. Immature oogonium showing papillae. $\times 813$.
- Fig. 20. Immature oogonia showing papillae.
- Fig. 21. Intercalary oogonium with three immature eggs. Note that sides of the eggs in contact are flattened. $\times 813$.
- Fig. 22. Oogonium with three immature eggs, antheridial stalk arising from lateral branch of oogonial stalk.
- Fig. 23. Irregular-shaped oogonium containing five eggs, the walls of which are becoming thickened.

PLATE 35

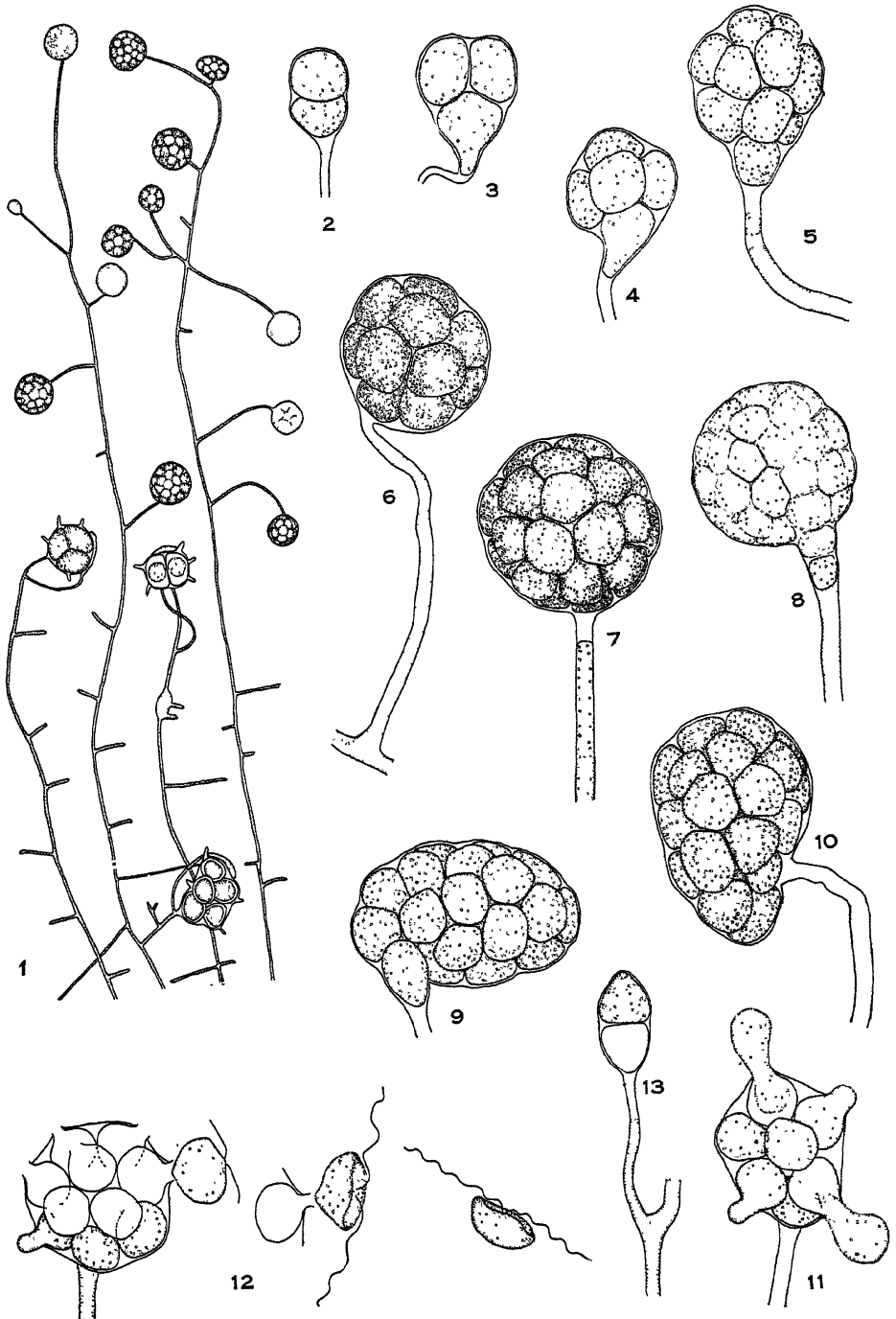
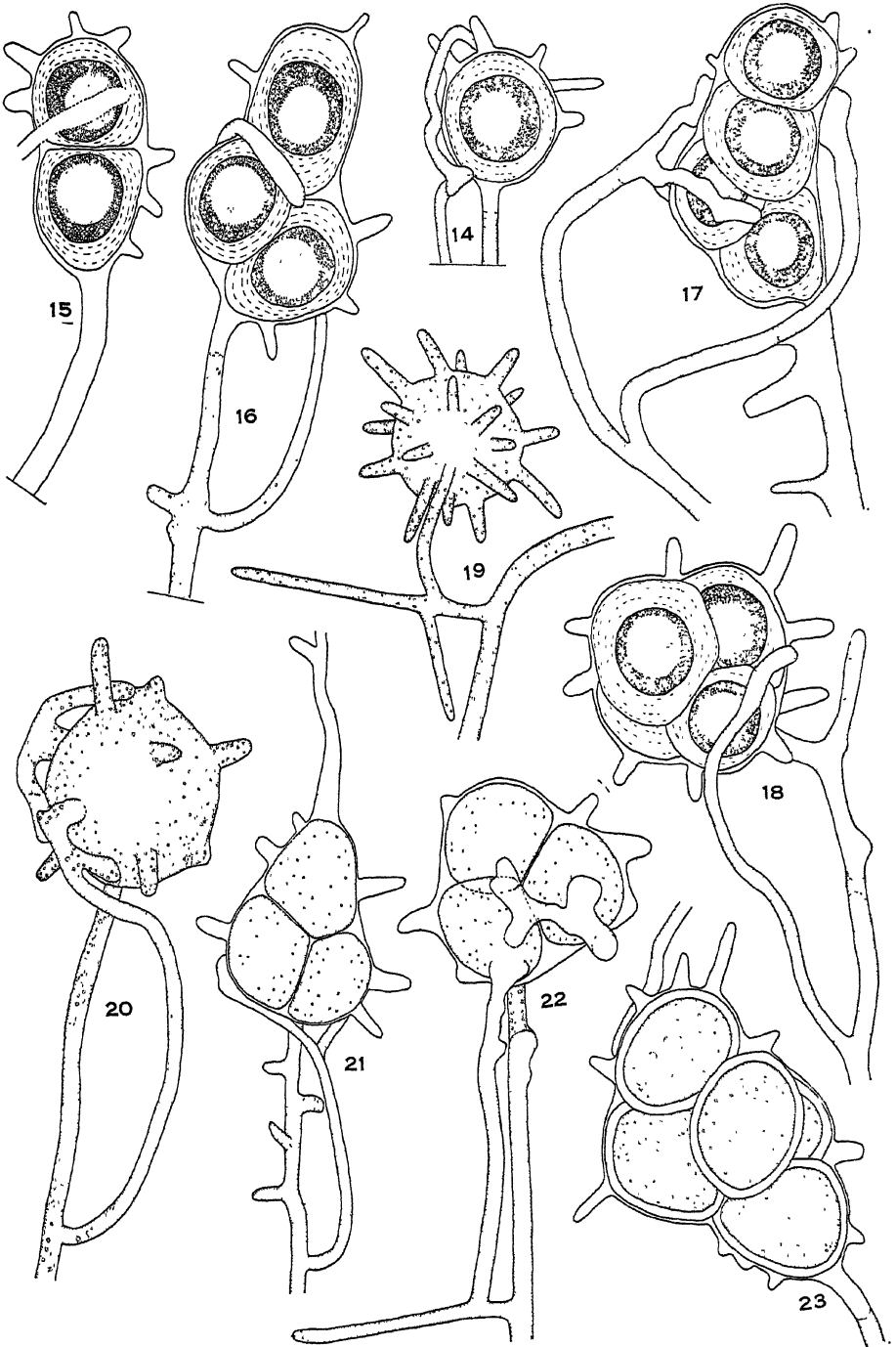


PLATE 36



NOTES ON CAROLINA FUNGI¹

By W. C. COKER

*Department of Botany, University of North Carolina
Chapel Hill, North Carolina*

PLATES 37-54

KEY TO FLESHY, STIPITATE POLYPORES OF NORTH CAROLINA

- Stem black below and with a black root.... . *P. radicans*
 Stem not black
 Tube surface rich wood brown; spores subspherical, 8-10 μ thick. *S. sublividus*
 Tube surface and entire plant blue(dull indigo or drab-blue); spores smooth, 3.7-4.4 x 4-4.8 μ *P. caeruleoporus*
 Tube surface white or light cream when quite fresh, then yellowish or greenish yellow or pale grayish
 Cap yellow, coarsely scaly; tubes and flesh turning green when rubbed ... *P. Ellisii*
 Cap brown, mottled with thin felted patches. *P. pes-caprae*
 Not as above
 Tubes very short(1-2 mm.) and small (3-4 to a mm.), their mouths smooth
 Cap smooth, grayish or straw color; no black layer beneath the tubes when dry; spores tuberculate, 4-5 x 5-6 μ *P. griseus*
 Cap nearly white, then pale orange yellow to cinnamon buff; usually with a black layer under the tubes when dry; spores smooth, 2.5-3.5 x 3.7-5 μ ... *P. ovinus*
 Tubes longer(2-6 mm.) and larger(1-2 to a mm.), their mouths jagged
 Cap greenish or brownish yellow, often cracked and areolated; spores 3.7-4.5 x 4-6 μ *P. cristatus*
 Cap buffy red or buffy ochraceous, not cracked; plant single; spores 3.5-4.2 x 5.5-7.5 μ *P. Stewartae*

Polyporus radicans Schw. Trans. Amer. Phil. Soc. II, 4: 155. 1832.

P. Morgani Frost in Peck. Rept. N. Y. St. Mus. 32: 34. 1879.

Scutiger radicans (Schw.) Murr. Bull. Torr. Bot. Club 30: 430. 1903.

Plates 37-39 and 53, fig. 1

Cap up to 10 cm. broad, nearly plane, velvety-tomentose, rimose-punctate toward the center, dry, ochraceous tawny to buffy leather color. Flesh thick, pure white, elastic, soft with the texture of marshmallow; tasteless and odorless.

Tubes pure white, dull brown when bruised, shallow, 2 mm. deep or less, about 0.5 mm. wide, walls thick when young, thinner and uneven when fully mature, irregularly decurrent, in some places descending to the black part of the stem.

Stem about 6-10 cm. long, contracting rather abruptly to a long, crooked, pliable, blackish root; stem terete above, uneven below, about 1.5-2.5 cm. thick in center, more or less unequal; surface velvety except where covered by the descending tubes, whitish or light ochraceous, abruptly changing below where covered by duff or leaves to deep blackish brown; solid, elastic, strong and tough, the flesh white.

Spores (of No. 14339) white, smooth, ellipsoid, 5.2-6.2 x 10-13.5 μ .

¹ Including some species of *Polyporus*, *Boletus*, *Lentinus*, *Armillaria*, and *Entoloma*.

When dry the plant is rigid and brittle, differing in this respect from any other species in the *Scutiger* section that we have studied.

In the young plants with tubes scarcely visible, as shown in our No. 14262, our notes are as follows: "Caps 3.5 cm. wide, strongly convex, velvety, not yet obviously tomentose, margin involute; color a deep rich red-brown, near Hessian brown (Ridg.). Tube surface at this time nearly white, tinted with salmon, staining red-brown when bruised, as does also the exposed stem." The roots in these plants were very large, much larger than the stem and very irregular. In all our collections the roots have been broken off before reaching the buried wood from which they are supposed to spring. In several of our plants the roots show branching at the base, as in Hard's figure.

Originally found in Pennsylvania by Schweinitz, this striking species has not been reported, so far as we know, from the south until Hesler found it in Tennessee (Jour. Tenn. Acad. Sci. 4: 16, 1929). We first collected it in 1917, but have not reported it until now. Murrill does not have it in his Southern Polypores (1915).

Illustrations: Hard. Mushrooms, fig. 329, 1908.

Lloyd. Syn. Sect. Ovinus, fig. 508. 1911. Also same in Syn. Stip. Polyp., fig. 465 bis (as *Ovinus radicans*). 1912.

Neuman. Bull. Wisc. Geol. and Nat. Hist. Survey, No. 33: pl. 15, fig. 54. 1914.

North Carolina. Chapel Hill. No. 2840. Low frondose woods (few scattered pines), Sept. 26, 1917. No. 7553. Mixed woods, Oct. 5, 1924. No. 13970. Mixed woods, Oct. 1, 1945. No. 14262. In a yard with pines near by, Oct. 9, 1947. No. 14339. By the stump of a deciduous tree, Battle Park, Oct. 28, 1947. No. 14350. Mixed woods, no stump near but not far from trees, Nov. 4, 1947.

Scutiger sublividus (Snell) Singer, Snell, and White. Mycologia 37: 128. 1945.

Polyporoletus sublividus Snell. Mycologia 28: 467. 1936.

Polyporus canaliculatus Overholts. Mycologia 33: 100. 1941.

Plates 40 and 53, figs. 2-4

Massive plants; caps up to 15 cm. broad and 13 cm. long, convex with reflexed edges, the thick sterile margin strongly inrolled; surface tomentose-squamulose with agglutinated fibers, bluish gray, becoming dull green then blackish when rubbed, the inturned margin soft-velvety, on exposure to rain becoming a sordid buff-brown and reticulated by collapsed fibers. Flesh pure chalk white, turning dull greenish or bluish in places on exposure, tough, homogeneous, like very firm marshmallow, not at all fibrous, 3-4 cm. thick at stem; taste faintly alkaline, odor none.

Tubes large to very large, covering the lower side of the horizontal stem all the way to the earth, smaller toward the margin (about 0.7 mm. wide there), those on stem up to 1.5-4 mm., intermingled with much smaller ones here and there, inner surface strongly glaucous, tube length about 5-12 mm., up to 15 mm. on the stem; flesh a deep soaked brown, their mouths a rich wood brown.

Stem almost lateral, massive, up to 3.5-4.5 cm. thick and 7 cm. long, subequal, the base flattened between roots; flesh just as in the cap; mycelium white.

Spores (good prints) pure chalk white, large, nearly spherical, apparently minutely punctate but really pitted, mucro sublateral, 7-8.5 x 7.5-9.7 μ . Basidia

4-spored, 11μ thick; sterigmata stout, about 7.5μ long. Threads of flesh rather freely branched, most about 7.5 – 13μ thick, with very thin walls; clamp connections conspicuous and many crystals on the threads.

This is a striking species, clearly marked by its blue-gray cap, handsome wood brown tube mouths, and large, subglobose spores with pitted walls. The tube color is not due to age, as it is the same in the developing margin as in mature areas. When dried the plants are very firmly tough and leathery, not brittle; the tubes are gray-brown with a tint of fawn, the cap drab with yellow and purple tints; the flesh distinctly tinted with pale salmon and with light red-brown blotches. In old plants, much exposed to rain, the cap surface loses its native color and becomes a sodden buffy brown. The inner surface of the tubes, when throwing spores, is strongly glaucous, appearing slatey blue. See Singer, Snell, and White (cited above) for a discussion of the spore wall structure.

Thanks to Dr. L. R. Hesler, we have seen parts of the types of *Polyporoletus sublividus* Snell and *Polyporus canaliculatus* Overholts, as well as other material from Tennessee mentioned by Singer, Snell, and White.

This species has been known heretofore only from Blount and Cocke counties in Tennessee.

North Carolina. Highlands. No. 12816. Emerging almost horizontally among exposed cut roots from a low bank, July 11, 1942. No. 13515. Same spot as above, Aug. 21, 1942. Also Hesler, No. 18670. Howell Forest, Aug. 31, 1948.

Polyporus caeruleoporus Peck. Rept. N. Y. St. Mus. **26**: 68. 1874.

Polyporus holocyaneus Atk. Journ. Mycol. **8**: 117. 1902.

Scutigera holocyaneus (Atk.) Murr. Bull. Torr. Bot. Club **30**: 429. 1903.

Scutigera caeruleoporus (Pk.) Murr. Bull. Torr. Bot. Club **30**: 429. 1903.

Plate 53, figs. 5, 6

Plant blue-drab all over; cap 2.4–10 (15) cm. wide, convex when young and usually with a little umbo which may or may not be visible at maturity, then unevenly plane, the center slightly depressed, margin membranous, sterile and strongly inrolled at all ages; surface dry, dull, minutely granular or roughened; color blue-drab (about deep madder-blue of Ridg. but a little more gray). Flesh solid, homogeneous, milk-white, just as in the stem; taste somewhat peppery, odor faint but rather distinctly fragrant, somewhat like spearmint.

Tubes short, (1) 2–5 mm., subcircular, 1–3 to a mm., larger and with mouths radially elongated near the stem, all the dissepiments irregular, of uneven height, and finely fimbriate-frosted; tubes more or less decurrent on one or more sides and in some cases all the way to the substratum, but much shallower on the stem except at times on the base when the stem is strongly slanting; color of cap but paler with a glaucous sheen, and becoming more gray and less blue at full maturity.

Stem equal or nearly so or somewhat enlarged downware, centric or excentric, crooked, color of cap, base where protected pure white with mycelium; flesh white, solid.

Spores (of No. 13100) white, subspherical, smooth, 3.7 – 4.4×4 – 4.8 . Basidia about 5.5 – 6μ thick, 4-spored.

In the south this is distinctly a mountain plant of cool damp hemlock-rhododendron woods. Our Highlands plants were growing by a cold rocky brook near the entrance to Calhoun Lodge. They were single or cespitose, often with tiny under-privileged little ones of different sizes coming up in contact with larger ones. The little ones are almost exactly like the larger ones, both in shape and in color. The context of the plants is firmly fleshy throughout but subject to early borings by grubs. The cap margin is usually more or less undulate or lobed. When dry these plants are softly elastic, not so tough as in the other members of this group that we have studied.

The species is now known from New York, Vermont, Pennsylvania, and from the mountains of North Carolina and Tennessee.

Illustrations: Coker. Journ. Elisha Mitch. Sci. Soc. **43**: pl. 22, fig. 9 (as *P. holocyaneus*, spores). 1927.

Hesler. Journ. Tenn. Acad. Sci. **16**: 172, fig. 9 (as *P. holocyaneus*). 1941.

North Carolina. Highlands. No. 13100. By a brook on the Dillard road, Sept. 2, 1942. No. 12298 (Hesler, coll.). Under hemlock and rhododendron, Aug. 29, 1939 (U.N.C. Herb.).

Linville Falls. Coker, coll.

Tennessee. Cades Cove. Hesler, No. 16587.

Polyporus Ellisii Berk. Grevillea **7**: 4. 1878.

Polyporus flavo-squamosis Underw. Bull. Torr. Bot. Club **24**: 84. 1897.

Scutiger Ellisii (Berk.) Murrill. N. Amer. Flora **9**: 64. 1907.

Plates 41, 42, and 53, fig. 15

Cap up to 24.5 cm. broad, laterally attached, often irregularly lobed and contorted, margin drooping and thin, surface coarsely fibrous and scaly, the scales being made by the cracking into areas of a dense superficial coat; color light yellow all over. Flesh thick, white, turning green when cut, taste very pleasant and mild, odor distinctly like spoiled ham.

Tubes rounded and rather regular, about $\frac{3}{4}$ mm. in diameter, white, but turning a beautiful sea green when rubbed, shallow, about 2-3 mm. deep and with thin partitions, somewhat decurrent, fading into reticulations towards the base.

Stem excentric, stout and short, expanding into a large irregular rooting base which may extend several inches into the ground and is connected with large yellow mycelial strands and plates.

Spores (of No. 892) white, smooth, ellipsoid, $5.5-6.5 \times 7.2-9.3\mu$.

This singular and rare plant has been found in only a few places (Alabama, South Carolina, New Jersey). It seems to prefer clay soil in pine woods.

Illustrations: Lloyd. Myc. Notes, Polyporoid Issue No. 2, fig. 264. Aug. 1909.

North Carolina. Chapel Hill. No. 610. In red clay in which there were pine roots, Oct. 22, 1912. Spores $5.1-6 \times 8.2-9\mu$. No. 638. On ground under pines, Oct. 26, 1912. No. 888. In clay bank by a road, Oct. 4, 1913. Spores white, ellipsoid, $5.5-7.4 \times 7.4-9.2\mu$. No. 892. In Battle Park, Oct. 8, 1913. No. 1472. Battle Park, dry

ground by path on hillside, Nov. 2, 1914. No. 1952. On a clay bank in pines, Oct. 31, 1915. No. 1999. Under pines, Nov. 21, 1915. No. 4671. In mixed woods, Oct. 10, 1920. No. 13254. In mixed woods, Nov. 5, 1942. No. 14361. At base of a stump, Nov. 8, 1947.

Polyporus pes-caprae (Pers.) Fr. Syst. Myc. 1: 354. 1821.

P. pes-caprae Pers. Champ. Comest., p. 241. 1818.

P. retipes Underw. Bull. Torr. Bot. Club 24: 85. 1897.

Scutiger retipes (Underw.) Murr. Bull. Torr. Bot. Club 30: 428. 1903.

Plates 43, 44 (bottom), and 53, fig. 16

Cap irregular, wavy and lobed, about 6–12 cm. broad, usually reniform or deeply notched at the lateral stem, roughened by thin, felted-tomentose, appressed patches, so as to appear finely mottled, becoming squamulose toward the thin margin; color a deep rich chestnut brown all over except for the paler cracks. Flesh thick and very firm and elastic, white, but grub channels red-brown; taste pleasant, odor none.

Tube surface chalk white, turning pallid (in No. 12452 turning olive green when rubbed); tubes decurrent, extending in some cases down the anterior half of the stem to the ground, shallow, about 2.5 mm. deep at best, large, up to 3–4 mm. wide, some smaller, very variable and irregular, in some places only partial plates and ridges, those on the stem either full length or reduced to wrinkles only.

Stem about 4–6 cm. long and 1.5–2 cm. thick, nearly equal, usually constricted at the ground to a short root or bunch of fibers, concolorous with cap except where covered by the tubes, roughened by small scales and patches, the very base yellowish; flesh very firm, solid, and elastic.

Spores white, smooth, ellipsoid, variable in the same print, 4.4–6.3 x 6–10.8 μ .

This interesting European plant was first recorded in America from Alabama by Underwood (as *P. retipes*, type). It is a rare species and seems to have been reported from only a few places (Alabama, Tennessee, North Carolina, and New Jersey; see Murrill, Mycologia 12: 9, 1920, and Hesler, Journ. Tenn. Acad. Sci. 20: 238, 1945).

Illustrations: Bresadola. Funghi Mang. et Vel., pl. 95 (as *P. scobinaceus*). 1899.

Gillet. Champ. Fr., Hymen., pl. 450 (566).

Lloyd. Myc. Notes No. 35: fig. 332. 1910.

Persoon. Champ. Comest., pl. 3.

Quélet. Champ. Jura et Vosg. 1: pl. 17, fig. 2.

Richon et Roze. Atl. Champ., pl. 62, figs. 5–8 (scales more conspicuous than in ours).

North Carolina. Chapel Hill. No. 881. In pine and oak woods, Oct. 5, 1913. No. 4870. Under pines, May 22, 1921.

Highlands. No. 2815. Bearpen Mountain (4200 ft.), Sept. 6, 1932 (Hesler). No. 9650.

On a road bank, Aug. 22, 1933. Spores pure white (print), smooth, 5.2–6.3 x 6–9.3 μ .

No. 9825. On a clay bank, Aug. 25, 1934. No. 11018. On a road bank in deciduous woods, Aug. 30, 1938. Spores 4.4–5.5 x 9.3–10.8 μ . No. 12452. By mountain trail, Cliffside, July 27, 1941. No. 12980 and 12981. On a road bank in deciduous woods, about 350 yards apart, Aug. 16, 1942.

Macon County. No. 10912. Rich deciduous woods, Coweeta Exp. Forest, Aug. 12, 1938.

Polyporus griseus Peck. Bull. Buf. Soc. Nat. Sci. **1**: 60. 1873.

Also in Rept. N. Y. St. Mus. **26**: 68. 1874.

Scutigera griseus (Pk.) Murr. Bull. Torr. Bot. Club **30**: 431. 1903.

Polyporus Earlei Underw. Bull. Torr. Bot. Club **24**: 84. 1897.

Plates 44 (top) and 53, figs. 7, 8

Plants single but gregarious; cap thick and heavy, very irregular and uneven, 6.5–12.5 cm. broad, often one-sided or deeply lobed, roughly expanded, convex to depressed in center; surface very smooth, shining, dull bluish gray to straw color or light brownish gray. Flesh very thick, up to 2 cm., firm, slightly bitterish-sweet, pure white or in center grayish brown with at times a tint of pink.

Tubes very shallow, less than one mm. deep in our plants, about 3–4 to a mm., pure white, then pale grayish in age, brown where bruised, only slightly decurrent, the mouths smooth.

Stem short and stout, about 2–4 cm. long and 2–4 cm. thick, often pointed below, nearly the color of the cap, solid and firm.

Spores coarsely nodulated, subglobose, 4–5 x 5–6 μ ; basidia 4-spored, 6 μ thick; cystidia none.

In the dried state the plant is extremely tough and not brittle; cap about the color of *ovinus* in spots but mostly darker and duller brown, and also with blackish areas if dried in poor condition; flesh buckthorn brown and without any black line beneath the tubes, which are themselves a soft buffy brown almost the same color as the flesh; stems not reddish but darker brown.

This species was originally described from New York and is now known from Alabama, Tennessee, North Carolina, and several northern states, including Montana in the west. It is easily distinguished by its dense, heavy habit, bluish gray color (over-all brown when dry), and sharply by its warted spores. We have never found the plant but once (a fine colony), and Lowe says it is rare in New York.

This species is very near if not the same as *P. leucomelas* (which also has nodulated spores) as illustrated by Fries (Icon. Hymen. **2**: pl. 179, fig. 1). Lloyd (Syn. Sect. Ovinus of Polyporus, p. 78, 1911) seems to think that they are the same. He refers to Boudier's plate 151 (Icon. Myc. **1**), as *P. leucomelas*, as an excellent illustration of *griseus*. This plate differs from our plants and also from Fries's plate in having a squamulose cap and stem. Murrill considers *P. Earlei* Underw. (Alabama) the same as *P. griseus* Pk., and from the description of the former we think he is right. We have not seen the type.

Illustrations: Lloyd. Syn. Sect. Ovinus of Polyporus, p. 78, figs. 498–500. 1911.

North Carolina. Chapel Hill. No. 3192. In low mossy place in deciduous woods, Oct. 5, 1918. A fine colony of 9 separate plants.

Polyporus ovinus (Schaeff.) Fr. Syst. Myc. **1**:346. 1821.

Boletus ovinus Schaeff. Icon. Fung., pls. 121, 122. 1763.

Scutigera ovinus (Schaeff.) Murr. Bull. Torr. Bot. Club **30**:426. 1903.

Plates 45, 46, and 53, figs. 9, 10

Medium to large, heavy, fleshy plants, single or cespitose, the stems often connate, growing on soil in pine or mixed woods; caps may be crowded and interlocked, 5–14 cm. broad, averaging about 10 cm., wavy and irregular, usually somewhat depressed in the center, the margin acute and more or less drooping; color when quite fresh whitish to pale straw on the smooth margin, shading to pale orange yellow or cinnamon buff toward the center, the superficial layer usually cracking into small or rather large, fibrous, darker patches (diffracted scaly). Flesh about 1 cm. thick at stem, thinning gradually to a sharp margin, white, soft, pliable, not brittle, turning yellow around grub channels and on drying pale brown or reddish brown. Often the grub channels are so numerous as to give the appearance of yellow to the whole flesh. Taste usually mild and pleasant but sometimes slightly bitter; odor faintly acid, becoming, in incipient decay, decidedly acid and attracting swarms of *Drosophila* flies.

Tubes very small, varying in size, about 3 to 5 to a mm., extremely shallow, only 1 mm. deep, descending the stem often almost to the base, white at first, changing to cream or clear light yellow, in fading changing to Pinard or empire yellow (Ridg.).

Stem more or less covered with the decurrent tubes, variable in size, usually 4–6 cm. long and 1.5–3 cm. thick, often pitted, channelled, tuberculate or flattened, usually somewhat swollen below; surface white, then cream or yellowish, deep red-brown when strongly bruised and base nearly always deep, dull red when dry; flesh like that of the cap.

Spores (of No. 13992) pure white, smooth, oval-ellipsoid, $2.5\text{--}3.5 \times 3.7\text{--}5\mu$. Basidia 4-spored, $5\text{--}6.5\mu$ thick; hymenium $22\text{--}26\mu$ thick. Threads of flesh hyaline, rather thin-walled, much contorted, variable in thickness, $\pm 2\mu$ or swollen in places up to 18μ thick, branched and with cross walls.

When dry *P. ovinus* is heavy, rigid, and very tough, not brittle; the cap color usually a reddish brown, as buckthorn or sayal brown (Ridg.), varying to paler brown, dull gray-brown, or even pallid gray in places. If dried when old or in incipient decay, the cap may in large part turn black. The tubes on drying vary greatly in color, more often becoming ochraceous or reddish brown or smoky buff, with or without a faint tint of olive. The tube bases as well as the thin layer of flesh that supports them are usually black in section. When this layer is exposed from below, by the eating away of the tubes by grubs, it has a very characteristic dark grayish slate color with a slight olive tint.

Polyporus ovinus is very near *P. confluens* and, if different, the distinctions made do not hold up well in our rather numerous collections. According to Overholts (Pa. St. Coll., Sch. Agric. Bull. **298**: 23, 26, 1933) and Lowe (N. Y. St. Coll. For. Bull. **60**: 36, 1942), the plant with reddish-brown caps (when dry) and habit often cespitose should be *confluens*. Our plants, more often than not, have these characters but also nearly always show the black line above the tubes when dry, a character confined to *ovinus* by Shope and Lowe. Since the publication of Lowe's paper, he has seen all our collections from Chapel Hill and has pronounced

them all *ovinus*. Shope considers *Scutiger laeticolor* Murr. and *S. Whiteae* Murr. the same as *P. confluens*. The illustration of *ovinus* given by Lloyd and reproduced by Shope does not look like our plant. Zeller (*Mycologia* 27: 459, 1935) reports *P. ovinus* from Oregon and mentions also collections from Alabama, Colorado, and New York.

Illustrations: Bresadola. *Icon. Myc.* 19: pl. 948.

Konrad and Maublanc. *Icon. Fung.* 5: pl. 421.

Schaeffer. *Icon. Fung.*, pl. 121 (good for our plants), pl. 122 (occasional in ours).

For the plant treated as *P. confluens*, see:

Kauffman. *Pap. Mich. Acad. Sci.* 1: pl. 34. 1921.

Shope. *Ann. Mo. Bot. Gard.* 18: pl. 28, fig. 1. 1931.

North Carolina. Chapel Hill. No. 914. In pine woods, forming an irregular fairy ring, Oct. 15, 1913. No. 941. In open woods south of athletic field, Oct. 18, 1913. No. 1338. In pine woods, Oct. 14, 1914. No. 1993. On ground in pine woods, Nov. 23, 1915. No. 3186. Under pines, Oct. 5, 1918. No. 9335. In mixed woods, Nov. 1, 1932. Spores $3-3.5 \times 3.4-4.2\mu$. No. 13992. Mixed woods near Forest Theater, Sept. 2, 1946. No. 14319. Mixed woods near Meeting of Waters, Oct. 20, 1947. Spores white, smooth, $2.5-3.5 \times 3.5-5\mu$. No. 14349. Mixed woods back of stadium, Nov. 4, 1947.

"Low and middle districts. Earth in woods." Curtis.

California. Trinidad, Humboldt County. *California Fungi* No. 502. H. E. Parks 4669, as *P. confluens*.

Polyporus cristatus (Pers.) Fr. *Syst. Myc.* 1: 356. 1821.

Boletus cristatus Pers. *Syn. Meth. Fung.*, p. 522. 1801.

Polyporus poripes Fr. *Nov. Symb.*, p. 48. 1851.

Polyporus flavovirens Berk. and Rav. *Ann. and Mag. Nat. Hist.* II, 12: 431. 1853. Also *Grevillea* 1: 38. 1872.

Grifola poripes (Fr.) Murr. *Bull. Torr. Bot. Club* 31: 335. 1904.

Plates 47, 48, and 53, fig. 11

A medium sized species with well defined but irregular and often confluent caps and stems; gregarious and often cespitose; compound masses may be up to 22 cm. broad. Caps expanded, uneven and interwoven, the thin margin in-rolled; surface velvety or glabrate, when young a rich yellowish brown, then greenish or yellowish tawny or paler, with grayish brown areas or zones, often cracked irregularly like the surface of dried mud, the cracks showing pale. Flesh whitish, up to 10-15 mm. thick, soft, toughish, often turning greenish when rubbed; when dry tough, sub-rigid and not very brittle, the plant retaining its form well.

Tubes white at first, then yellowish or greenish yellow, about 0.5-1 mm. wide and up to 3-5 mm. deep, the mouths angular and irregularly notched and toothed; strongly decurrent, sometimes to the ground. On drying the tubes in young plants turn a lively rosy pink, or cinnabar color, usually less so as they approach maturity, but even at full maturity the tubes may dry reddish.

Stem usually about 3-7 cm. long and 1-2 cm. thick, tough, solid, often more or less contorted and fused with others, usually concolorous with cap where not covered by the descending tubes, but often paler.

Spores (of No. 1868) white, smooth, subglobose to ovoid, $3.7-4.5 \times 4-6\mu$, with one droplet.

This is distinctly a fall species in the piedmont south. It is very resistant to decay and in Chapel Hill may be found in good condition into November, usually on rich slopes in deciduous woods. It is not at all rare in central and western North Carolina, but seems to be much rarer in the northern states. The type of *P. poripes* was from North Carolina, collected by Dr. M. A. Curtis.

When fresh this plant is tough and pliable, not woody. It is edible but rather too tough unless prepared by special recipes. Konrad and Maublanc (cited below) say that sulfuric acid gives the flesh a pretty violet color. We have two collections from Chapel Hill (Nos. 1805 and 16088) in which the tubes were aborted, leaving the hymenial surface minutely rugose, and no spores being formed. Microscopic examination showed no infesting fungus.

Most keys to species of *Polyporus* are misleading in placing this plant only under "stem branched" or "compound," as a colony is apt to have as many simple individuals as compounds ones. Lowe keys it under both branched and unbranched groups, which saves confusion.

Illustrations: Barla. Champ Nice, pl. 29, figs. 4-7. 1859.

Bresadola. Icon. Myc. 20: pl. 971.

Hard. Mushrooms, fig. 327 (as *P. flavovirens*). 1908.

Konrad and Maublanc. Icon. Sel. Fung. 5: pl. 423.

Krombholz. Abbild. u. Besch., pl. 48, figs. 15, 16.

Lloyd. Syn. Sect. Ovinus of Polyporus, fig. 501. 1911.

Overholts. Wash. Univ. Stud. 3: pl. 1, fig. 5, a, b, 1915.

Rostkovius in Sturm. Deutschl. Flora 3: fasc. 5, pl. 16.

Roumeguère. Crypt. Illus., fig. 215.

Schaeffer. Icon. Fung., pl. 113 (as *Boletus flabelliformis*).

North Carolina. Chapel Hill or near. Fourteen collections taken from 1908 to 1944 in the months of September and October in rich deciduous woods in humus or on mossy clay banks.

Highlands. Eight collections in August and September from same habitat as in Chapel Hill.

Haywood County. Pink Bed Valley. No. 10978. Aug. 23, 1938.

Linville Falls. No. 5739. Aug. 24, 1922.

Macon County. Coweeta Exp. Forest, Aug. 1938

Blowing Rock. Atkinson, coll. (as *P. flavovirens*).

Winston-Salem. Schallert, coll. On old stump, Sept. 10, 1922.

Reported by Schweinitz (as *Boletus cristatus*), presumably from Salem.

"Middle and upper districts, wooded ravines." Curtis (as *P. poripes*).

Polyporus Stewartae n. sp.

Plates 49 (top) and 53, figs. 12-14

Plant solitary, thick and heavy, drying thin and light; cap irregularly expanded, 5-11.5 cm. wide, the margin thin, fertile; surface buffy red (pinkish cinnamon, Ridg.), turning purplish red when bruised, very smooth, glabrous and dull like leather. Flesh thick, about 3.5 cm. at stem margin and descending into it with little change, creamy white, rather firm but tender; taste slight, mildly bitter, odor not strong but distinct, rather unpleasantly aromatic; after standing some time bruised flesh turning about the same color as the bruised tubes.

Tubes up to 6 mm. long, decurrent and shorter toward stem, where their mouths are much elongated radially, a few ending in delicate reticulations on the stem; color light cream, becoming *brown* when bruised; surface uneven, mouths irregular, about 1-1.5 mm. wide except toward the stem, edges more or less jagged.

Stem thick, firm, up to 5 cm. long and 3 cm. thick in center, expanding into the cap and tapering downward to a blunt, shallowly inserted base; surface glabrous, color of tubes but dark brown on exposure or rubbing, solid, flesh pale yellow.

Spores (type) light yellow (hyaline under microscope), oval-ellipsoid, more flat on one side and sometimes slightly broader near the proximal end, $3.5-4.2 \times 5.5-7.5\mu$. Basidia 4-spored, $6.7-7.5\mu$ thick and about 40μ long; sterigmata slender, straight, about 5μ long. Cells of both cap and tube flesh very variable in width and length, cross walls abundant, no clamps seen, walls very thin and easily collapsing.

When dry the plant is sub-rigid but not brittle, extremely tough, almost like leather, the cap surface much wrinkled and of a bright orange buff color, between orange buff and ochraceous buff (Ridg.), with or without pinkish blotches, the surface of both tubes and stem a dull buffy brown, about clay color (Ridg.), cut surfaces between cream-buff and chamois, with a silky sheen.

Distinguished from others nearest it by solitary habit, bright-colored, perfectly glabrous cap, rather long and large pores, and bright color when dry.

Named for Miss Josephine Stewart, who collected the type.

North Carolina. Chapel Hill. No. 85. In woods near cemetery, Oct. 10, 1913. Spores smooth, $3.7-4.5 \times 5.5-7.5\mu$. No. 13989 (type). Deciduous woods, south of campus, Oct. 4, 1945.

Polyporus balsameus Peck. Rept. N. Y. St. Mus. **30**: 46. 1878.

P. crispellus Peck. Rept. N. Y. St. Mus. **38**: 91. 1885.

Plate 54, fig. 1

Plants small, thin, applanate, petaloid, and narrowing to a point, confluent and imbricated; individual caps about 12 mm. to 4.5 cm. wide and up to 5 cm. long, white with narrow concentric zones of brownish gray, fibrous-tomentose.

Tubes very minute, about 1.5 mm. deep, white at first, then speckled with brown, becoming more brown when drying.

Flesh pure white, about 1.5 mm. thick in central region, much thicker near point of attachment, very tough, strongly acid-bitter, drying rigid.

Spores (of No. 9613) small, hyaline, oval-ellipsoid, smooth, $2-2.5 \times 3.5-4\mu$. (Lowe gives $3-5 \times 2-3\mu$.)

Found on the wood of coniferous trees, causing a brown rot. So far as we know, it has previously been reported from the south only by Hesler from the mountains of North Carolina (Highlands?) (Mycologia **26**: 196, 1934) and Tennessee (Castanea **2**: 51, 1937).

Illustrations: Hubert. Phytopathology **19**: 725, figs. 1-3. 1929. Also Outl. For. Path., 345, fig. 89. 1931.

McCallum. Bull. Canada Dept. Agric. n.s. **104**: pl. 7, figs. 2, 3. 1928.

North Carolina. Highlands. No. 9036 (det. by Murrill). Aug. 3, 1932. Spores $1.8-2.8 \times 3.7-4.5\mu$. No. 9613 (det. by Overholts). On hemlock wood by Ravenel Lake, Aug. 15, 1933.

Polyporus rhipidium Berk. Dec. Fungi No. 124, Hooker's Lon. Journ. Bot. 6: 319. 1847.

Favolus rhipidium (Berk.) Cke. Grevillea 15: 58. 1886.

Plates 49 (bottom) and 54, fig. 2

Gregarious in dense groups in imbricated rows, the stalks crowded and sometimes fused at base, or some plants single, distinctly stalked laterally, convex or almost plane, not umbilicate, up to 2.4 cm. wide and 1.5 cm. long, more or less reniform, vaguely zonate, smooth but with a felted appearance, pale straw color when fresh, nearly white when dry; margin even, thin, somewhat deflexed. Flesh homogeneous, up to 2 mm. thick, soaked straw color when fresh; flesh of stem similar, solid. Taste at first mild, soon strongly peppery, contracting the throat and making the saliva mucilaginous as with slippery elm; odor none.

Tubes up to 0.8 mm. long, about 3-4 to a mm. in least diameter, the mouths angular, radially much elongated toward the margin, nearly isodiametric toward the stem, concolorous; pore surface nearly even.

Stem up to 1.2 cm. long, often shorter but always distinct, terete or somewhat grooved or flattened, color and surface like cap but base slightly scurfy.

Spores (of No. 10282) pure white, minute, ellipsoid, smooth, $2.2-2.8 \times 3.7-4.5\mu$.

The plants are strongly hygrophanous and very quickly revive in water. Dried plants are nearly pure white on cap and stem, but tube color nearly unchanging; rather chalky brittle when dry, pliable but cheesy and not tough when damp.

This is a widely distributed but apparently rare species. So far as we know, it has been reported from the southern states only by Ravenel from South Carolina and Curtis from North Carolina. It is known in the United States from New England, New York, and Ohio, also from the tropics of both hemispheres and from New Zealand.

Illustrations: Cunningham. New Zealand Polyporaceae. 3. The Genus *Polyporus*, pl. 2, fig. 5. 1948.

Lloyd. Polyp. Issue No. 2, p. 23, fig. 260, a-e. 1909. See also his Polyp. Issue No. 3, p. 33. 1910.

Neuman. Polyporaceae of Wisconsin, p. 32, pl. 1, fig. 7 (Wisc. Geol. and Nat. Hist. Survey Bull. 33: 158).

South Carolina. Darlington. No. 10282. On bark of a rotting red maple stump by Swift Creek, Sept. 16, 1936.

Myrtle Beach. No. 13722. On rotting decorticated stub of wax myrtle, July 28, 1946. Spores white, $2-2.5(3) \times 3.7-4.6\mu$.

Boletus Ananas Curtis. Amer. Jour. Sci., ser. 2, 6: 351. 1848.

B. isabellinus Peck. Bull. Torr. Bot. Club 24: 146. 1897.

Plates 50 (bottom) and 54, fig. 3

Since our note, with illustration, in this Journal (64: 142, pl. 23, top) we have had the opportunity to observe more closely the habits of this interesting species. At the State Park a few miles below Myrtle Beach, S. C., in flat, sandy, mixed woods (pine, oak, dogwood, etc.) we found ten plants and on the Platt estate, a few miles farther south, two more. Of these twelve, nine were attached to living pine trees (in all but one case only one to a tree), but up to three inches above the ground, arising from a more or less obvious mat of white mycelium, the trees showing no sign of rot; one plant was in open ground about 15 feet from pines; and one was on the ground under an exotic azalea bush. All previous reports where specific data are noted give its association with pine and it seems very likely from what we now know that the species is a mycorrhizal symbiont of the pine. Its habits suggest that in an attempt to elevate the sporophore the mycelium climbs the nearest tree, which would usually be its host, but if the pine is too far away it will climb some other tree or do without a tree if necessary.

We are now adding another photograph of one of our recent collections (No. 14434). Curtis mentioned the appendiculate cap margin, a conspicuous feature, but no one has described it as a veil. In our accompanying plate, the small plant shows the veil just as it is breaking from the stem. It is a thick, tough, fibrous membrane, all of which remains as torn fragments on the cap margin.

Peck's *B. isabellinus* is the same as the present species, as given by Murrill (N. Amer. Flora 9: 156), and as admitted, after some discussion, by Snell (Mycologia 25: 229-231; Publ. I of R. I. Bot. Club, p. 20). We have studied the spores of Peck's type and find them exactly as in our plants, 6-8 x 14.8-18 (19) μ , with distinct longitudinal striae. From Peck's description one would not suspect that the plants are the same. His spore measurements are much too short. The hymenium of our plant (No. 14434) is about 30-35 μ thick, dark brown, contrasting strongly with the hyaline context below it; basidia ovate with a basal neck, 15 μ thick, 37 μ long, with 4 short, stout sterigmata; cystidia present but not abundant, rounded below and with a long beak projecting up to 40 μ .

Lentinus tigrinus (Bull.) Fr. Syst. Myc. 1: 176. 1821.

Agaricus tigrinus Bull. Herb. Fr., pl. 70.

Lentodium squamulosum Morgan. Journ. Cin. Soc. Nat. Hist. 18: 36. 1895.

Plates 50 (top) and 54, fig. 5

Cap about 1-5 cm. broad, umbilicate with an inrolled margin, in age becoming infundibuliform, surface yellowish brown, covered with deep brown or blackish scales and fibers which are more numerous in the center, and in youth often make the whole surface very dark. Flesh very thin, white, said to become red at

times when wounded, tough, tasteless and odorless. A delicate veil may or may not be obvious.

Gills crowded, white then yellowish, decurrent, serrate or eroded, veined at the cap and at times anastomosing to some extent to form irregular pores.

Stem centric or excentric, 1-5 cm. long, 2-5 mm. thick in middle, curved, tapering downward in large plants, squamulose-fibrous, color of cap, firm, solid, tough.

Spores (of No. 81) white, smooth, ellipsoid, $3-3.8 \times 7-9\mu$.

Rather rare on rotting stumps and logs of frondose trees.

For a good treatment of this and other species, see Harper (Trans. Wisc. Acad. Sci. **20**: 365. 1922). He finds this species up to 3 inches but so far we have it only up to 5 cm. Some European illustrations also show plants larger and stouter than ours.

Lentinus tigrinus is often attacked and deformed by a fungus parasite which may completely cover the gills with a white blanket. *Lentodium squamulosum* is a name given by Morgan to this form, considering it an autonomous aborted variant and not the result of a parasite. This view was also advocated by Lyman (Boston Soc. Nat. Hist. **33**: 181, pl. 23) who grew the aborted form from spores. Harper thinks Lyman's cultures not convincing and that the abnormality is probably due to a species of *Hypomyces*, a very reasonable conclusion. Both normal and abnormal forms are often found in the same colony, as in our No. 5881.

Illustrations: Bresadola. Icon. Myc. **11**: pl. 509.

Bulliard. Herb. Fr., pl. 70.

Cooke. Ill. Brit. Fungi, pl. 1138.

Harper. Trans. Wisc. Acad. Sci. Arts and Let. **20**: pls. 18-20, 21, figs. A-D. 1922.

Moffat. Nat. Hist. Surv. (Chicago Acad. Sci.) Bull. **7**: pl. 6, fig. 2. 1909.

Morgan. Journ. Cin. Soc. Nat. Hist. **18**: pl. 1, fig. 3 (*Lentodium*). 1895.

Patouillard. Tab. Anal., fig. 406.

Ricken. Blätterpilze **1**: pl. 26, fig. 2.

Rolland. Atl. Champ., pl. 45, fig. 97.

North Carolina. Chapel Hill. No. 81. On stump of an apple tree, Oct. 28, 1911. No. 3127. On a very rotten stump of frondose tree, New Hope swamp, May 5, 1918. No. 3760. Parasitized form. On a decaying stump, Nov. 16, 1919. No. 5881. On a decaying gum stump, Oct. 8, 1922. All but one of these 12 plants had the gills parasitized. No. 7050. On a deciduous log, Aug. 5, 1923. Spores(print) white, smooth, ellipsoid, $3.4-3.8 \times 7.4-9.3\mu$.

Lentinus Ravenelii B. & C. Hooker's Journ. Bot. & Kew Misc. **1**: 100. 1849.

Plates 51 (top) and 54, fig. 6

Cap 1.3-4.2 cm. broad, deeply umbilicate, the margin usually drooping, surface inconspicuously squamulose, the small squamules almost inherent and concolorous, more numerous in the center, the surface being nearly white except in the umbilicus where it is light brown; margin somewhat undulate, in young plants strongly incurved and with a tendency to split in drying. Flesh thin, very tough and elastic; taste and odor fungoid.

Gills subdistant, a little decurrent, interveined, 2.5-3.7 mm. wide, the margins delicately serrate.

Stem short, slender (1–2 cm. x 1.5–2.5 mm.), slightly enlarged above and below, central or more often excentric, surface like that of the cap, solid, very firm and tough.

Spores (of No. 44, good prints) cream color, ellipsoid with a small excentric mucro, smooth, $2.9\text{--}3.5 \times 5.5\text{--}7.7\mu$.

Plants long dried absorb water very slowly, but overnight they expanded almost to their original shape.

That this is *L. Ravenelii* seems almost certain. It is a small plant with a slender stem and is evidently related to *L. tigrinus* but its size, delicacy, very pale color and somewhat smaller spores justify a name, although possibly it should be considered a variety of *tigrinus*. Ravenel found his plants on the Santee (lower South Carolina) "On decaying wood in wet places," which was just the habitat of our plants. In some cases fine threads could be seen between the gills, but there was no conspicuous parasitism, and a number of good spore prints were thrown. *Lentinus Dunali* mentioned by Harper (Trans. Wisc. Acad. 20: 375) does not look like our plant (See Berkeley, Outl. Brit. Fung., pl. 15, fig. 2).

South Carolina. Hartsville. No. 17. Abundant on a sweetgum log on creek below the lake, June 4, 1917. Plants like No. 44 except a little darker. Spores $2.5\text{--}3.3 \times 4.4\text{--}7\mu$. No. 44. On blackgum logs on margin of the lake, Aug. 7, 1917.

Lentinus sulcatus Berk. Lond. Jour. Bot. 4: 301. 1845. (Decades of Fungi VIII, No. 77)

Plates 51 (bottom) and 54, fig. 7

Cap 1.3–1.8 cm. broad, convex, not umbilicate, beautifully sulcate half way to center, inherently fibrous, minutely squamulose, the center cracked into larger scales; color tawny, the center darker. Flesh tough, pliable, pale, about 1 mm. thick near stem.

Gills rather close, adnexed, about 1.5 mm. wide, thick, the margin lightly notched, color pale cream.

Stem central, 1.1–1.4 cm. long, 1.2–1.5 mm. thick above, tapering a little downward, fibrous-lined, furfuraceous above, tough, curved, brown below, pale above.

Spores (of No. 4630) cream color when fresh, smooth, oval-ellipsoid or oblong, $6\text{--}7.8 \times 11\text{--}15\mu$; after years in the herbarium spore print pinkish ochraceous.

Growing from cracks in very hard wood, as telephone poles, arbor posts, etc. (all our collections on coniferous wood). *Lentinus pholiotoides* Ellis and And. (Bot. Gaz. 16: 45, 1891), which is supposed to be the same, grew on dead *Populus*. If wet again a few days after drying our plant will revive, and if in good condition cast another spore print.

Panus fulvidus Bres. (Fungi Trid. 2: 56, pl. 165; Icon. Myc. 11: pl. 518), a European species, while a larger form, is too near to be easily separated. Our American plant seems to have been found heretofore only in the north and west, and so far we have seen no illustration of it.

North Carolina. Chapel Hill. No. 4445. On a telephone pole, July 19, 1920. No. 4630. On a telephone pole, Aug. 6, 1920. No. 4868. On a cedar post, May 12, 1921.

Armillaria caligata Viviani. Funghi Ital., p. 40. 1834.

Agaricus (*Armillaria*) *nardosmius* Ellis. Bull. Torr. Bot. Club 6: 75. 1876.

Armillaria nardosmia (Ell.) Sacc. Syll. Fung. 5: 86. 1887.

Plates 52 and 54, fig. 4

Plants large, handsome and solid; cap up to 13 cm. wide, surface smooth or with small scales in center, somewhat silky-shining and mottled and lined with appressed fibers and scales, not viscid; color a peculiar shading of reddish brown and violaceous or grayish tan. Margin strongly involute in youth, extending beyond the gills and continuous with the large fibrous veil which is thinnest at the margin. Flesh firm, grayish white, about 12 mm. thick near center; taste bitterish (very obviously peppery in No. 14028), odor none in our form.

Gills crowded, deepest in middle, up to 1 cm. wide, pure white to grayish white in youth, becoming brownish in age, slightly notched at stem.

Stem 4-6 (10) cm. long, up to 2 cm. thick, equal or somewhat enlarged below, smooth and color of cap below the veil, white and delicately fibrous or granular above; texture fibrous and elastic, the center fibrous-stuffed; base of stem at times badly eroded by grubs and in these cases almost black.

Veil very large, a thin strong membrane colored like the cap on the outside and attached about 2.5-3.5 cm. from the base of the stem and clothing the lower part of the stem with the same tissue; at the margin the fibers becoming separated in part and leaving short fragments attached to the edge of the cap. When not too much torn the ring stands out like an ample ruff on the stem but later collapses.

Spores pure white (fresh), turning yellowish, short-ellipsoid, smooth, 3.7-5.5(6) x 6-8.5(9) μ , with a large oil drop.

We are in agreement with Smith in considering this species the same as Ellis's *Agaricus nardosmius*. Under both of these names the plant has been described as having a distinct odor or no odor: that of *caligata* variously given as of radish, pears, or "fruity"; of *nardosmia* as of spikenard, almonds, or "disagreeable." Smith (Pap. Mich. Acad. Sci. Arts and Let. 19: 206. 1934) describes three forms of *caligata* according to odor or lack of it: (1) *caligata*, typical, odor fruity; (2) forma *inodora*; (3) forma *nardosmia*, odor disagreeable. By this test all of our North Carolina plants would be the *inodora* form.

Until recently this species has been confused in this country with *Agaricus robustus* Alb. & Schw. (Consp. Fung., p. 147. 1805). Our plant can hardly be other than what Schweinitz reported as frequent in woods in our territory as *Agar. (Lepiota) robustus* (Syn. Fung. Car. No. 561. 1822). As *Armillaria caligata* had not then been described and is very closely related it is not surprising that he should refer the North Carolina plant to his European species. Other early reports, as by McIlvaine, were as *robusta*. Kauffman describes what he considers the true *robusta* from the Adirondack Mountains, N. Y. (Pap. Mich. Acad. Sci. 2: 64, pl. 8. 1922). It differs from *caligata* in more reddish (rufous)

cap and stem below the veil, tendency of the flesh of cap and stem to become reddish, and smaller and less globose spores. Kauffman had previously fully described *caligata* in The Agaricaceae of Michigan (p. 650). For European descriptions and comparisons of these two species, see Boudier (Icon. Myc. 4: 11) and Bresadola (Funghi Mang., pp. 30-31; Icon. Myc. 1: pls. 44, 45).

Illustrations: Boudier. Icon. Myc. 1: pl. 21.

Bresadola. Funghi Mang., pl. 17; Icon. Myc. 1: pl. 45.

Dufour. Atl. Champ., pl. 8, fig. 15 (as *A. robusta* var. *caligata*).

Hard. Mushrooms, fig. 42 (as *A. nardosmia*).

Kauffman. Agaricaceae of Michigan, pl. 137.

Rolland. Atl. Champ. Fr., pl. 13, fig. 21.

Smith. Pap. Mich. Acad. Sci. Arts and Let. 19: pls. 36, 37. (1933) 1934.

North Carolina. Chapel Hill. No. 826. In thick underbrush in Battle Park, Sept. 24, 1913. Spores 4-5.5 x 6-8.3 μ . No. 835. On ground among leaves, Sept. 25, 1913. No. 898. In leaves in mixed woods, Battle Park, Oct. 3, 1913. No. 1387. In woods, Battle Park, Oct. 17, 1914. Spores subglobose to ellipsoid, 4-6 x 5.2-8.5 μ . No. 9885. In rich deciduous woods, Oct. 8, 1934. Spores 4.2-5.2 x 6.5-8 μ . No. 13220. Deciduous woods by old Hillsboro road, Oct. 25, 1942. A slender form. Spores oval with oblique mucro, 4.3-5 x 6-8 μ .

Highlands. No. 12191. On bank by a road, Aug. 20, 1939. Spores 4-5(6) x 6-8(9) μ .

Tryon. Deciduous woods, Nov. 9, 1936. D. C. Peattie, coll. Spores 4.2-5 x 6.8-8 μ .

South Carolina. Hartsville. No. 12330. In Kalmia Gardens, Dec. 23, 1940. Spore 4.4-5.6 x 5.5-7(8) μ .

Note: We have received from Dr. L. R. Hesler a good collection of *A. caligata* from Cades Cove, Tennessee (No. 16545), that in the dried state has a very distinct and pleasing fragrance that might be called "fruity." Following Smith, this should be considered typical. Hesler had previously reported *caligata* from Blount County, Tenn. (Journ. Tenn. Acad. Sci. 12: 242. 1937).

Entoloma cuspidatum Pk.

Plate 54, fig. 8

Cap conical-campanulate with a small prominent papilla, viscid, glabrous; texture fibrous, easily splitting radially; margin uneven. No odor.

Gills pale yellow, then with a tint of flesh over the yellow (not bright flesh color as in Kauffman), 3-4 mm. broad and broadly adnate, rather distant, margin uneven.

Stem long, slender, glabrous, color of cap, toughish and pliable, hollow, nearly equal, loosely attached with white mycelium.

Spores pink, irregular, usually more or less cubic with the mucro on one angle, 7.4-9.4 x 9.4-11.2 μ . Basidia clavate, 4-spored, about 11 μ thick near tip.

In drying the color fades to almost white and the papilla splits and shrinks to almost nothing.

The smaller plants in Kauffman's Agaricaceae, pl. 110, are good examples of our form.

North Carolina. Durham. No. 14475. Mixed woods on grounds of Duke University, Aug. 13, 1948. Jean Leitner, coll.

EXPLANATIONS OF PLATES

PLATE 53

- Fig. 1. *Polyporus radicans*. No. 14339. Spores.
Figs. 2, 3, and 4. *Scutiger sublividus*. No. 12816. Spores, one basidium, and threads of flesh showing crystals.
Figs. 5, 6. *Polyporus caeruleoporus*. No. 13100. Habit sketch and bit of hymenium.
Figs. 7, 8. *Polyporus griseus*. No. 3192. Spores and bit of hymenium.
Figs. 9, 10. *Polyporus ovinus*. No. 13992. Threads of flesh and spores.
Fig. 11. *Polyporus cristatus*. No. 1868. Spores.
Figs. 12, 13, and 14. *Polyporus Stewartae*. No. 13989. Spores, hymenial cells, and cells of flesh, respectively.
Fig. 15. *Polyporus Ellisii*. No. 892. Spores.
Fig. 16. *Polyporus pes-caprae*. No. 4870. Spores.
All spores $\times 1620$; figs. 3, 6, 8, 13 $\times 810$; fig. 5 $\times \frac{1}{4}$.

PLATE 54

- Fig. 1. *Polyporus balsameus*. No. 9613. Spores, one beginning to sprout.
Fig. 2. *Polyporus rhipidium*. No. 13722. Spores.
Fig. 3. *Boletus Ananas*. No. 14434. Basidia, cystidia, and two sterile cells.
Fig. 4. *Armillaria caligata*. No. 826. Spores.
Fig. 5. *Lentinus tigrinus*. No. 7050. Spores.
Fig. 6. *Lentinus Ravenelii*. No. 44. Spores.
Fig. 7. *Lentinus sulcatus*. No. 4445. Spores.
Fig. 8. *Entoloma cuspidatum*. No. 14475. Spores.
All spores $\times 1620$; fig. 3 $\times 810$.

10

•

"I'm not,"

5 0 0 0

1. 2. 3. 4. 5. 6. 7. 8. 9. 10. 11. 12. 13. 14. 15. 16. 17. 18. 19. 20. 21. 22. 23. 24. 25. 26. 27. 28. 29. 30. 31. 32. 33. 34. 35. 36. 37. 38. 39. 40. 41. 42. 43. 44. 45. 46. 47. 48. 49. 50. 51. 52. 53. 54. 55. 56. 57. 58. 59. 60. 61. 62. 63. 64. 65. 66. 67. 68. 69. 70. 71. 72. 73. 74. 75. 76. 77. 78. 79. 80. 81. 82. 83. 84. 85. 86. 87. 88. 89. 90. 91. 92. 93. 94. 95. 96. 97. 98. 99. 100. 101. 102. 103. 104. 105. 106. 107. 108. 109. 110. 111. 112. 113. 114. 115. 116. 117. 118. 119. 120. 121. 122. 123. 124. 125. 126. 127. 128. 129. 130. 131. 132. 133. 134. 135. 136. 137. 138. 139. 140. 141. 142. 143. 144. 145. 146. 147. 148. 149. 150. 151. 152. 153. 154. 155. 156. 157. 158. 159. 160. 161. 162. 163. 164. 165. 166. 167. 168. 169. 170. 171. 172. 173. 174. 175. 176. 177. 178. 179. 180. 181. 182. 183. 184. 185. 186. 187. 188. 189. 190. 191. 192. 193. 194. 195. 196. 197. 198. 199. 200. 201. 202. 203. 204. 205. 206. 207. 208. 209. 210. 211. 212. 213. 214. 215. 216. 217. 218. 219. 220. 221. 222. 223. 224. 225. 226. 227. 228. 229. 230. 231. 232. 233. 234. 235. 236. 237. 238. 239. 240. 241. 242. 243. 244. 245. 246. 247. 248. 249. 250. 251. 252. 253. 254. 255. 256. 257. 258. 259. 260. 261. 262. 263. 264. 265. 266. 267. 268. 269. 270. 271. 272. 273. 274. 275. 276. 277. 278. 279. 280. 281. 282. 283. 284. 285. 286. 287. 288. 289. 290. 291. 292. 293. 294. 295. 296. 297. 298. 299. 300. 301. 302. 303. 304. 305. 306. 307. 308. 309. 310. 311. 312. 313. 314. 315. 316. 317. 318. 319. 320. 321. 322. 323. 324. 325. 326. 327. 328. 329. 330. 331. 332. 333. 334. 335. 336. 337. 338. 339. 340. 341. 342. 343. 344. 345. 346. 347. 348. 349. 350. 351. 352. 353. 354. 355. 356. 357. 358. 359. 360. 361. 362. 363. 364. 365. 366. 367. 368. 369. 370. 371. 372. 373. 374. 375. 376. 377. 378. 379. 380. 381. 382. 383. 384. 385. 386. 387. 388. 389. 390. 391. 392. 393. 394. 395. 396. 397. 398. 399. 400. 401. 402. 403. 404. 405. 406. 407. 408. 409. 410. 411. 412. 413. 414. 415. 416. 417. 418. 419. 420. 421. 422. 423. 424. 425. 426. 427. 428. 429. 430. 431. 432. 433. 434. 435. 436. 437. 438. 439. 440. 441. 442. 443. 444. 445. 446. 447. 448. 449. 450. 451. 452. 453. 454. 455. 456. 457. 458. 459. 460. 461. 462. 463. 464. 465. 466. 467. 468. 469. 470. 471. 472. 473. 474. 475. 476. 477. 478. 479. 480. 481. 482. 483. 484. 485. 486. 487. 488. 489. 490. 491. 492. 493. 494. 495. 496. 497. 498. 499. 500. 501. 502. 503. 504. 505. 506. 507. 508. 509. 510. 511. 512. 513. 514. 515. 516. 517. 518. 519. 520. 521. 522. 523. 524. 525. 526. 527. 528. 529. 530. 531. 532. 533. 534. 535. 536. 537. 538. 539. 540. 541. 542. 543. 544. 545. 546. 547. 548. 549. 550. 551. 552. 553. 554. 555. 556. 557. 558. 559. 560. 561. 562. 563. 564. 565. 566. 567. 568. 569. 570. 571. 572. 573. 574. 575. 576. 577. 578. 579. 580. 581. 582. 583. 584. 585. 586. 587. 588. 589. 590. 591. 592. 593. 594. 595. 596. 597. 598. 599. 600. 601. 602. 603. 604. 605. 606. 607. 608. 609. 610. 611. 612. 613. 614. 615. 616. 617. 618. 619. 620. 621. 622. 623. 624. 625. 626. 627. 628. 629. 630. 631. 632. 633. 634. 635. 636. 637. 638. 639. 640. 641. 642. 643. 644. 645. 646. 647. 648. 649. 650. 651. 652. 653. 654. 655. 656. 657. 658. 659. 660. 661. 662. 663. 664. 665. 666. 667. 668. 669. 670. 671. 672. 673. 674. 675. 676. 677. 678. 679. 680. 681. 682. 683. 684. 685. 686. 687. 688. 689. 690. 691. 692. 693. 694. 695. 696. 697. 698. 699. 700. 701. 702. 703. 704. 705. 706. 707. 708. 709. 710. 711. 712. 713. 714. 715. 716. 717. 718. 719. 720. 721. 722. 723. 724. 725. 726. 727. 728. 729. 730. 731. 732. 733. 734. 735. 736. 737. 738. 739. 740. 741. 742. 743. 744. 745. 746. 747. 748. 749. 750. 751. 752. 753. 754. 755. 756. 757. 758. 759. 760. 761. 762. 763. 764. 765. 766. 767. 768. 769. 770. 771. 772. 773. 774. 775. 776. 777. 778. 779. 780. 781. 782. 783. 784. 785. 786. 787. 788. 789. 790. 791. 792. 793. 794. 795. 796. 797. 798. 799. 800. 801. 802. 803. 804. 805. 806. 807. 808. 809. 810. 811. 812. 813. 814. 815. 816. 817. 818. 819. 820. 821. 822. 823. 824. 825. 826. 827. 828. 829. 830. 831. 832. 833. 834. 835. 836. 837. 838. 839. 840. 84

PLATE 37



Polyporus radicans. No. 2840. \times about $\frac{3}{4}$.

PLATE 38



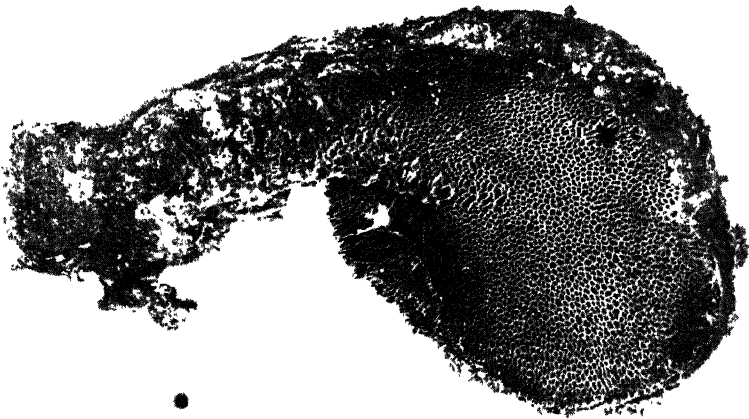
Polyporus radicans (dried). No. 14350. Slightly reduced.

PLATE 39



Polyporus radicans (dried). No. 14262. Very young plants. Nat. size.

PLATE 40



Scutiger sublividus. No. 13515. \times about $\frac{3}{4}$.

PLATE 41



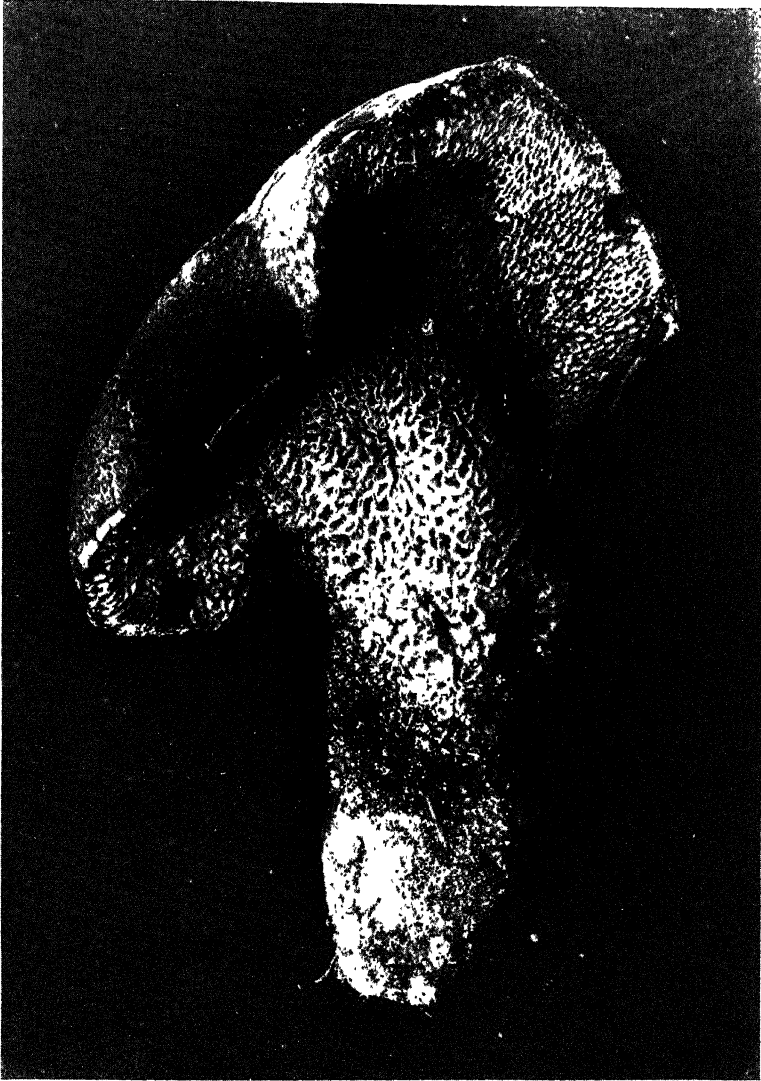
Polyporus Ellisii. No. 892. Slightly reduced.

PLATE 42



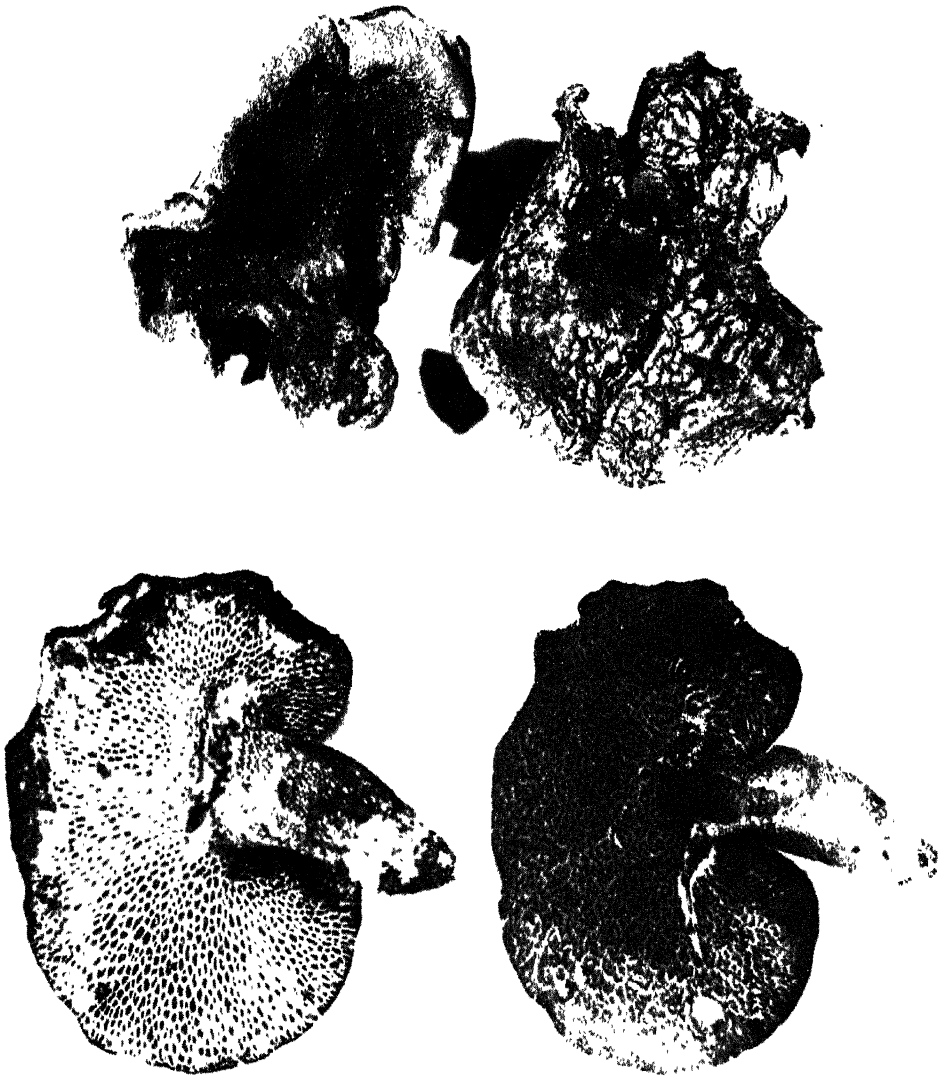
Polyporus Ellisii. No. 638. \times about $\frac{1}{2}$.

PLATE 43



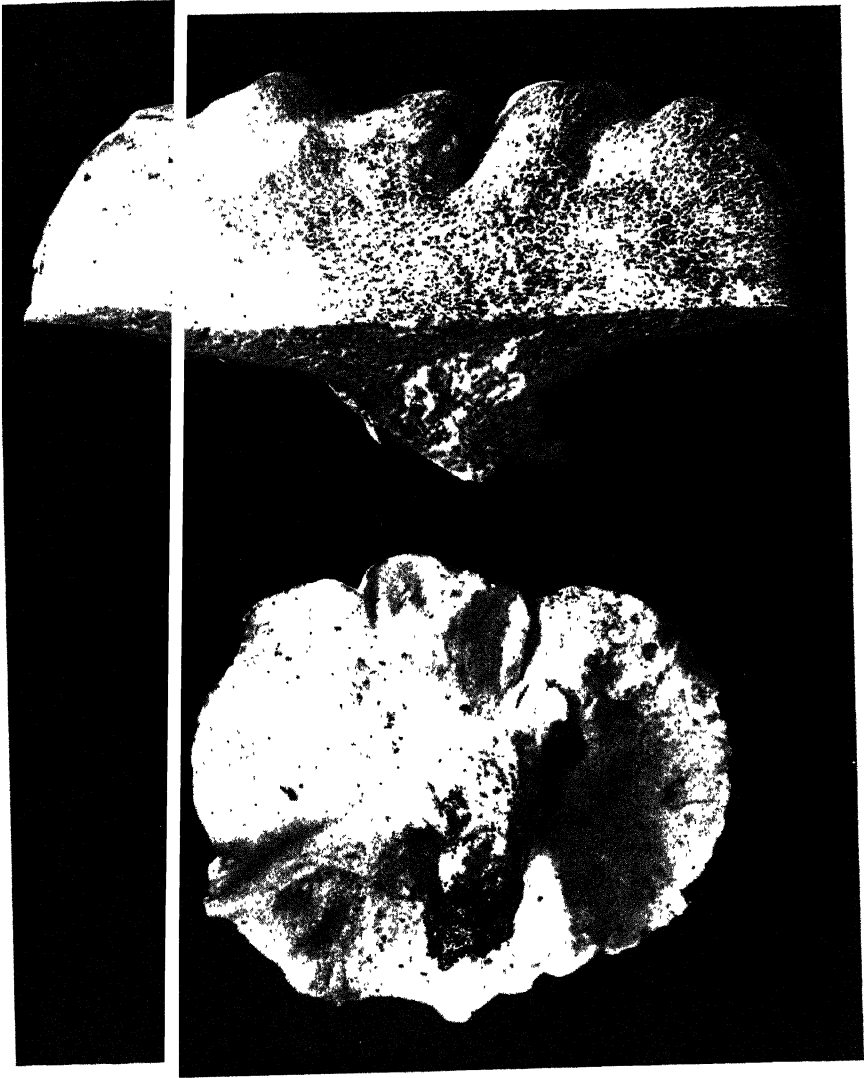
Polyporus pes-caprae. No. 4870. Nat. size.

PLATE 44



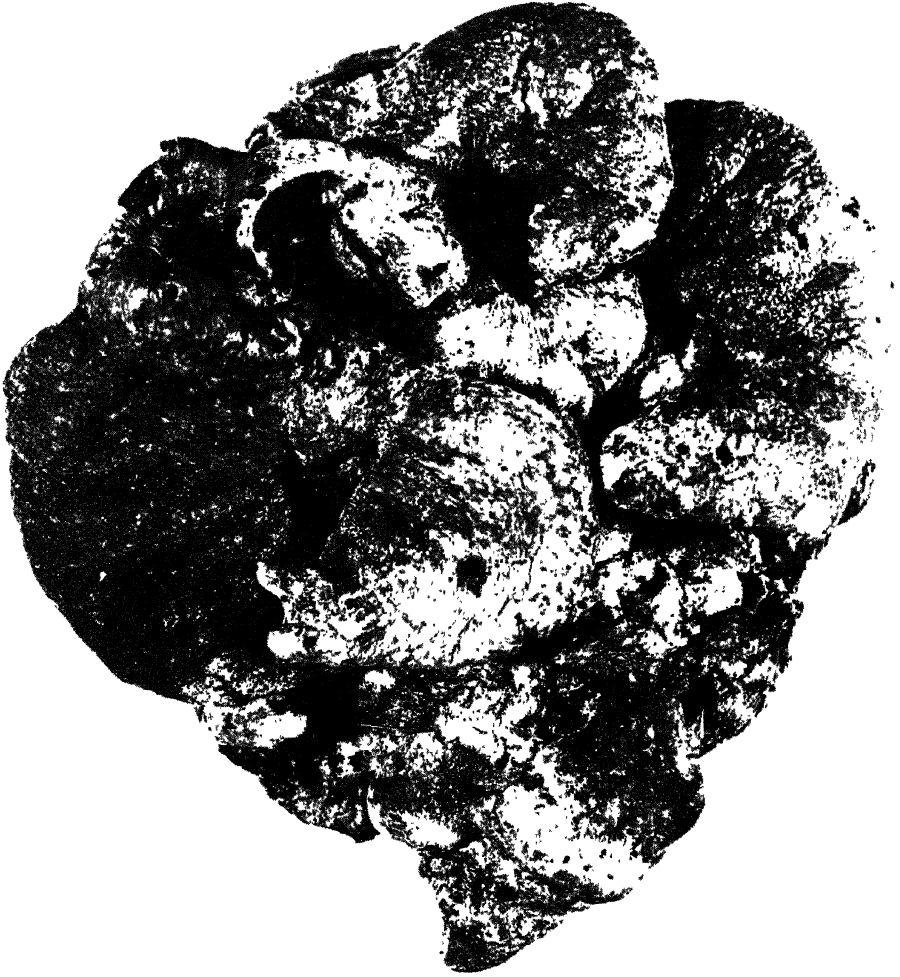
Polyporus griseus (top, dried plants). No. 3192.
Polyporus pes-caprae (bottom). No. 12452. Nat. size.

PLATE 45



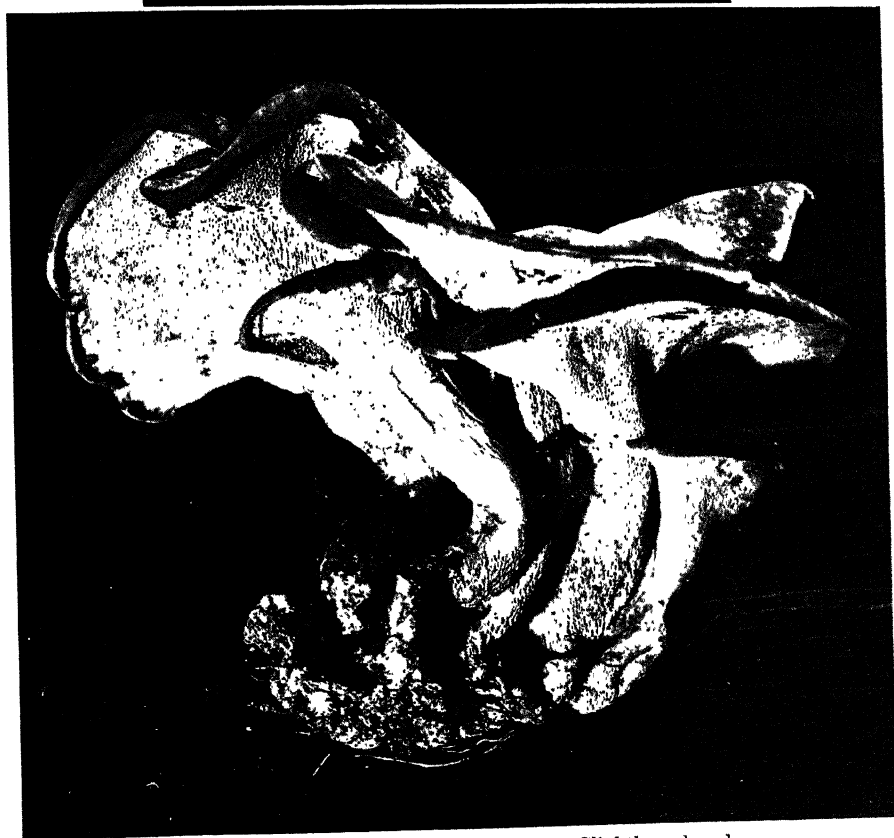
Polyporus ovinus. No. 3186. Slightly reduced.

PLATE 46



Polyporus ovinus (dried cluster). No. 914. Slightly reduced.

PLATE 47



Polyporus cristatus. No. 13619 (top). Slightly reduced.
No. 14240 (bottom, dried plants). Nat. size.

PLATE 48



Polyporus cristatus. No. 859. $\times \frac{5}{8}$

PLATE 49



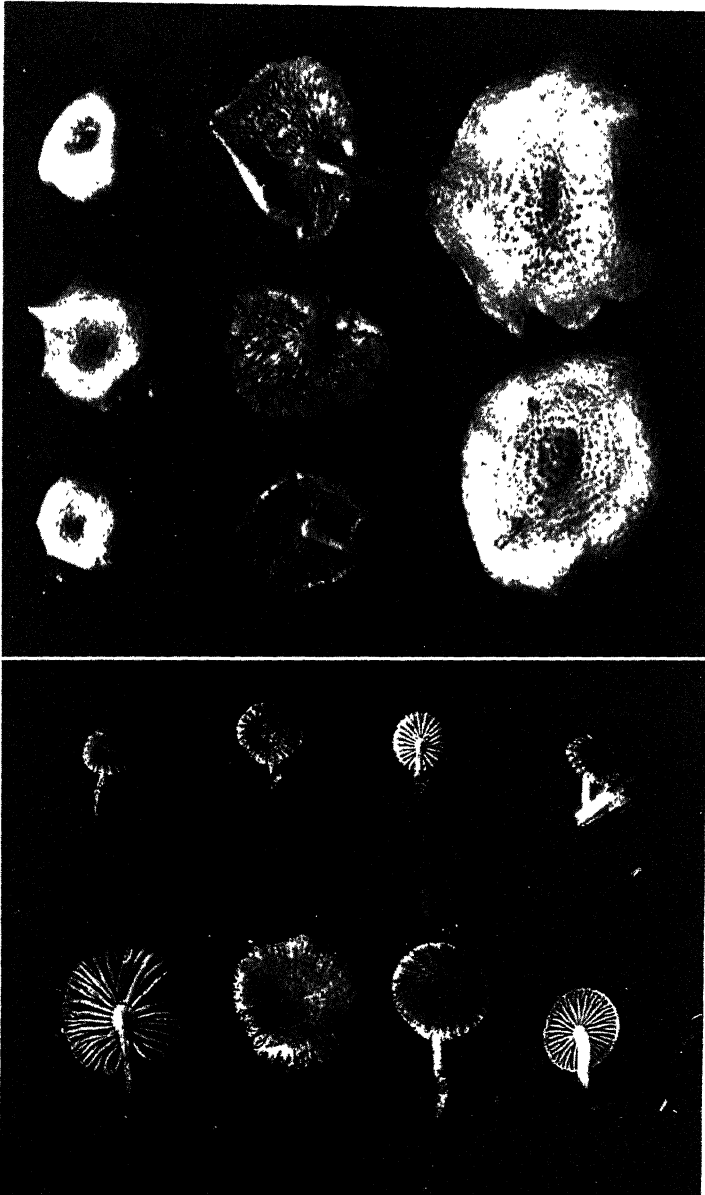
Polyporus Stewartae (top). No. 13989. \times about $\frac{3}{4}$.
Polyporus rhipidium (bottom, plant rewet from dried state). No. 10282. \times about $\frac{1}{4}$.

PLATE 50



Lentinus tigrinus (top). No. 3027. Nat. size.
Boletus Ananas (bottom). No. 14434. Nat. size.

PLATE 51



Lentinus Ravenelii (top). No. 44. Slightly reduced.
Lentinus sulcatus (bottom). No. 4630. Nat. size.

PLATE 52



Armillaria caligata forma *inodora*. No. 835 and (inset) No. 9885. Nat. size.

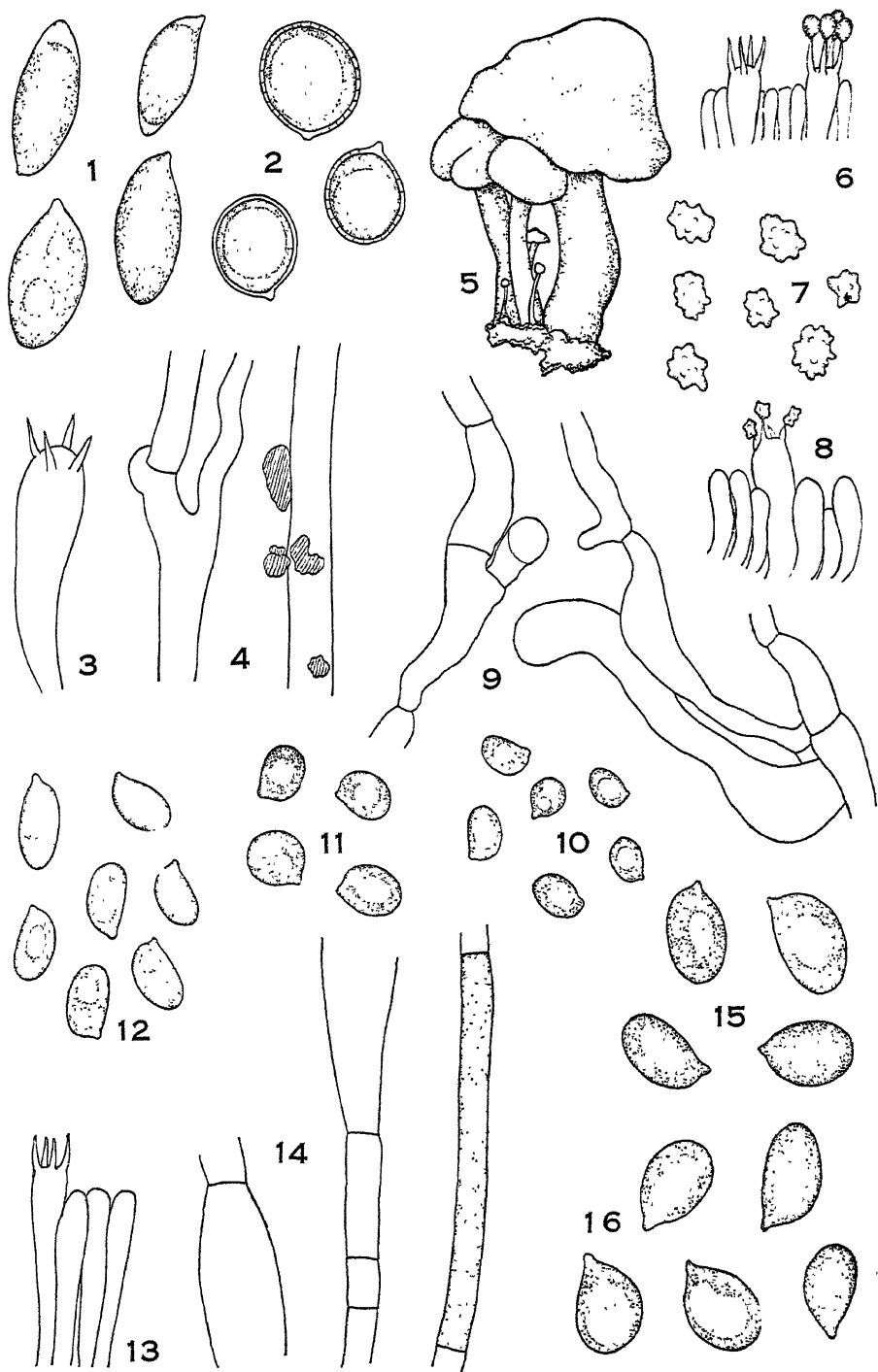


PLATE 54



I. A. R. I. 75.

IMPERIAL AGRICULTURAL RESEARCH
INSTITUTE LIBRARY
NEW DELHI.

[illegible]